VALUE OF C-REACTIVE PROTEIN ESTIMATION IN CONSERVATIVE MANAGEMENT OF P.R.O.M.

THESIS
FOR
DOCTOR OF MEDICINE

(OBSTETRICS AND GYNAECOLOGY)

M.L.B. MEDICAL COLLEGE

BUNDELKHAND UNIVERSITY,

JHANSI (U.P.)

2004 DEEPALI

CERTIFICATE

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This is to certify that the work entitled, **"Value of C-reactive protein estimation in conservative management of P.R.O.M"** which is submitted as a thesis for **M.D.** (Obstetrics and Gynaecology) by **Deepali singh**, has been carried out in the **Department of Obstetrics and gynaecology**, M.L.B. Medical college, Jhansi.

She has put in the necessary stay in the department as per university regulation.

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Dated: *Dr. Deepali Singh*

CONTENTS

INTRODUCTION

INTRODUCTION

The management of patients with premature rupture of membranes (PROM) poses one of the most serious dilemmas in obstetrics since PROM significantly increases the likelihood of prematurity and serious perinatal infection. PROM often results in preterm labor followed by a preterm delivery. The incidence of preterm delivery is estimated to be around 10 percent of all pregnancies, leading to 75 to 90 percent of perinatal morbidity and mortality. It has been proposed that intrauterine infection is one of the causes of preterm labor (10 to 40 percent). Signs of infection--fever, leucocytosis, elevated ESR--are absent or appear late. Chorioamnionitis (infection involving the chorion, amnion, and amniotic fluid, and usually the placental villi and decidua as well) occurs in 6 percent of preterm deliveries without PROM, and in 27 percent with PROM, resulting in a fourfold increase of neonatal mortality. PROM may be associated with 20 percent of all perinatal deaths. Obstetricians must choose whether to deliver the baby or to maintain the pregnancy for as long as possible, knowing that the second option may increase the risk for fetal infection. Reliable diagnosis of early infection could lead to better monitoring of PROM patients and a decrease in perinatal morbidity and mortality. So early detection of chorioamnionitis is of utmost importance in management of PROM. Hence a reliable predictor of infection was sought for and that is C-reactive protein. C-reactive protein is an acute phase reactant produced by hepatocytes. It raises significantly following injury and inflammation. Because of this latter association, it was thought that it could aid in the management of PROM. CRP may be

used in clinical practice to predict infection resulted from PROM during conservative management. CRP was found the most reliable indicator of histologic chorioamnionitis and indicated the presence of intrauterine infection earlier than WBC or ESR.

Premature rupture of membranes

During pregnancy, the fetus is surrounded and cushioned by a liquid called amniotic fluid. This fluid, along with the fetus and the placenta, is enclosed within a sac called the amniotic membrane. The amniotic fluid is important for several reasons. It cushions and protects the fetus, allowing the fetus to move freely. The amniotic fluid also allows the umbilical cord to float, preventing it from being compressed and cutting off the fetus's supply of oxygen and nutrients. The amniotic membrane contains the amniotic fluid and protects the fetal environment from the outside world. This barrier protects the fetus from organisms (like bacteria or viruses) that could travel up the vagina and potentially cause infection.

Although the fetus is almost always mature at between 36-40 weeks and can be born without complication, a normal pregnancy lasts an average of 40 weeks. At the end of 40 weeks, the pregnancy is referred to as being "term." At term, labor usually begins. During labor, the muscles of the uterus contract repeatedly. This allows the cervix to begin to grow thinner (called effacement) and more open (dilatation). Eventually, the cervix will become completely effaced and dilated. In the most common sequence of events (about 90% of all deliveries), the amniotic membrane breaks (ruptures) around this time. The baby then leaves the uterus and enters the birth canal.

Ultimately, the baby will be delivered out of the mother's vagina. In the 30 minutes after the birth of the baby, the placenta should separate from the wall of the uterus and be delivered out of the vagina.

Sometimes the membranes burst before the start of labor, and this is called premature rupture of membranes (PROM). There are two types of PROM. One occurs at a point in pregnancy before normal labor and delivery should take place. This is called preterm PROM. The other type of PROM occurs at 36-40 weeks of pregnancy.

<u>INCIDENCE OF PROM</u>: Gunn GC, Mishell DR Jr, Mortan DG (1970) found that the incidence of PROM ranges from 2-18%. Reports of Polansky GH et al (1985) showed the incidence PROM ranges from 14-17%. Overall, about 10% of all gestations are complicated by PROM. At term, the incidence of PROM varies from 6 to 19%.

<u>RISK FACTORS FOR PROM:</u> The causes of PROM have not been clearly identified. Some risk factors include **smoking**, multiple pregnancies (twins, triplets, etc.), and excess amniotic fluid (polyhydramnios). Certain procedures carry an increased risk of PROM, including **amniocentesis** (a diagnostic test involving extraction and examination of amniotic fluid) and cervical cerclage (a procedure in which the uterus is sewn shut to avoid **premature labor**). **placental abruption** is also associated with PROM, although it is not known which condition occurs first. In some cases of preterm PROM, it is believed that bacterial infection of the amniotic membrane causes it to weaken and then break. There is evidence to suggest that when the membranes are stressed, either by internal

pressure due to labor or by infection, they are weakened and have an increased susceptibility to premature rupture. Examples of some organisms commonly associated with PROM include bacterial vaginosis, Trichomonas vaginalis , mycoplasmae, chlamydia trachomitis , Neisseria gonnorhea , group B Streptococci . Other risk factors are incompetent cervix, poor nutritional status, Non-white race, Multiparity, Low pregnancy weight gain, Recent coitus, Advanced maternal age.

According to Lni (1984) antenatal vaginal examination is associated with premature rupture of membranes.

SIGNIFICANCE OF PROM: Because most patients with PROM deliver within 48 hours of membrane rupture, the significance of PROM depends on the gestational age of the fetus at its occurrence. With expectant management, approximately 9 of 10 term patients will progress spontaneously into labor with a latency period of no more than 48 hours. The complications that may follow PROM include premature labor and delivery of the fetus, infections of the mother and/or the fetus, and compression of the umbilical cord (leading to oxygen deprivation in the fetus).

Labor almost always follows PROM, although the delay between PROM and the onset of labor varies. When PROM occurs at term, labor almost always begins within 24 hours. Earlier in pregnancy, labor can be delayed up to a week or more after PROM. The chance of infection increases as the time between PROM and labor increases. The types of infections that can complicate PROM include amnionitis and endometritis. Amnionitis is an infection of the amniotic membrane. Endometritis is an infection of the innermost

lining of the uterus. Amnionitis occurs in 0.5-1% of all pregnancies. In the case of PROM at term, amnionitis complicates about 3-15% of pregnancies. About 15-23% of all cases of preterm PROM will be complicated by amnionitis. The presence of amnionitis puts the fetus at great risk of developing an overwhelming infection (sepsis) circulating throughout its bloodstream. Preterm babies are the most susceptible to this life-threatening infection. One type of bacteria responsible for overwhelming infections in newborn babies is called group B streptococci. The consequences of PROM for the neonate fall into three major overlapping categories.

1. Significant neonatal morbidity and mortality associated with prematurity
2. Complications during labor and delivery that increase the risk for neonatal resuscitation
3. Infection

C-reactive protein

Human C-reactive protein (CRP) is the classical acute phase reactant, the circulating concentration of which rises rapidly and extensively in a cytokine-mediated response to tissue injury, infection and inflammation. C-reactive protein (CRP) has been a measure of acute phase reactions to inflammation for the last 15 years. Recently improved high sensitive and standardized quantitative assays in serum and cerebrospinal fluid (CSF) have allowed a re-evaluation of its potential as a diagnostic laboratory test. CRP is an abnormal serum glycoprotein produced by the liver during acute inflammation. Because it disappears rapidly when inflammation subsides, its detection signifies the presence of a

current inflammatory process. Further, by serial measurements important information can be obtained on the resolution or continuation of the inflammatory process.

C-reactive protein was first described by Tillet and Francis in 1930. They concluded that sera of patients suffering from acute infection precipitated with a non-proteic pneumococcus extracts called C polysaccharide in the presence of calcium ions. The protein that caused this reaction was therefore called C-reactive protein (CRP). All acute inflammatory processes (infectious and non-infectious), and certain malignant conditions, result in rise in serum CRP as a non-specific phenomenon. CRP production is a non-specific response to disease and it can never, on its own, be used as a diagnostic test. However if the CRP result is interpreted in the light of full clinical information on the patient, then it can provide exceptionally useful information.

CRP is exclusively made in the liver and is secreted in increased amounts within 6 hours of an acute inflammatory stimulus. The plasma level can double at least every 8 hours, reaching a peak after about 50 hours. After effective treatment or removal of the inflammatory stimulus, levels can fall almost as rapidly as the 5-7-hour plasma half-life of labelled exogenous CRP. The only condition that interferes with the "normal" CRP response is severe hepatocellular impairment.

CRP can bind to a number of molecules, including phosphate esters, lipids, polyanions (DNA polylysin), polycations (histones, protamine) and a variety of polysaccharids. CRP is synthesized by the liver under regulatory control of cytokines. Synthesis of CRP and

other acute phase proteins by hepatocytes is modulated by cytokines. Interleukins 1b and 6 and tumour necrosis factor are the most important regulators of CRP synthesis.

FUNCTIONS OF CRP: The function of CRP is felt to be related to its role in the innate immune system (Du Clos, Terry V, 2000). Similar to immunoglobulin IgG, it activates *complement*, binds to Fc receptors and acts as an opsonin for various pathogens. Interaction of CRP with Fc receptors leads to the generation of proinflammatory cytokines that enhance inflammatory response. Unlike IgG, which specifically recognizes distinct antogenic epitopes, CRP recognizes altered self and foreign molecules based on pattern recognition. Thus CRP is thought to act as a surveillance molecule for altered self and certain pathogens. This recognition provides an early defence and leads to a proinflammatory signal and activation of the humoral, adaptive immune system.

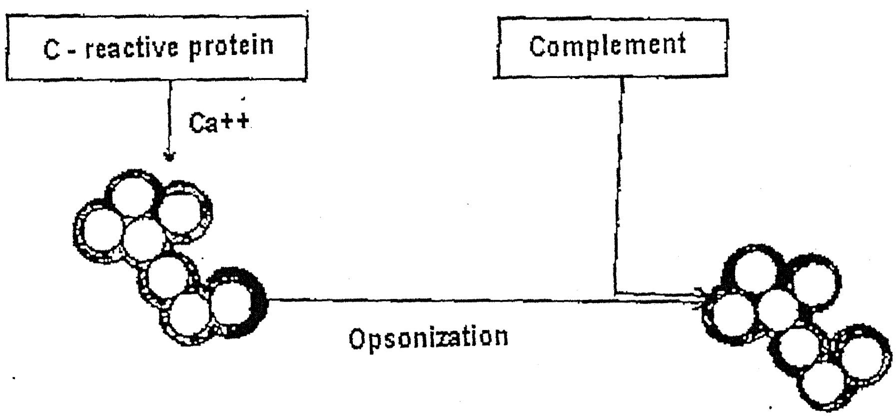


Fig.1 CRP binds to molecular groups found on a wide variety of bacteria and act as an opsonin.

Thus a number of functions have been ascribed to CRP, including initiation of opsonization and phagocytosis and activation of complement (Fig.1), neutrophils, and monocyte-macrophage. Collectively these properties imply an important role for CRP in the recognition of microbial organisms and as an immunomodulator in the host defence. CRP may also be important in the recognition of necrotic tissues.

WHY MEASURE CRP?

Levels of CRP increase very rapidly in response to trauma, inflammation and infection and decrease rapidly with the resolution of the condition. Since an elevated CRP level is always associated with pathological changes, determination of CRP is of great value in diagnosis, treatment and monitoring of inflammatory conditions. CRP is a more sensitive and reliable indicator of inflammatory processes than the ESR and the leucocyte count. The serum CRP concentrations increase faster than that of the ESR and when the condition subsides, CRP falls very quickly, reaching normal levels several days before the ESR normalises.

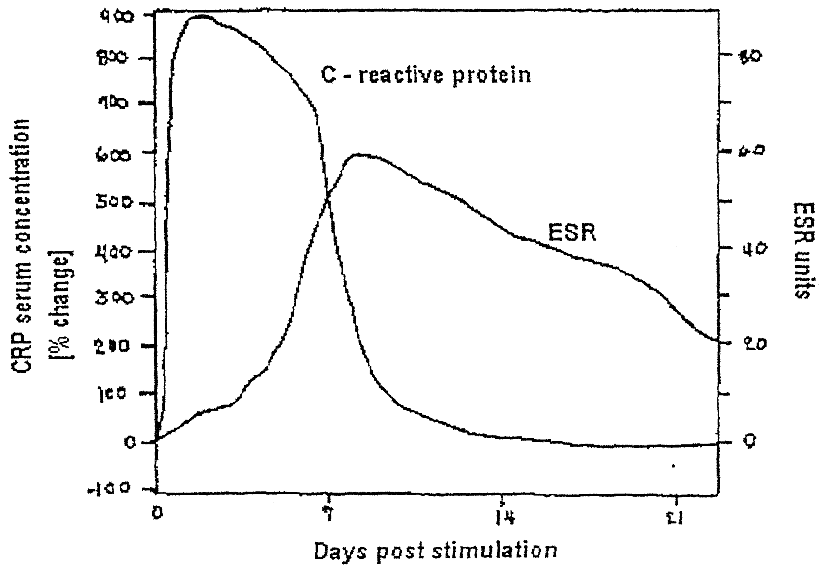


Fig. 2 CRP begins to rise in bacterial infections within 4-6 hours, peaks at 36-50 hours, closely parallels acute response with 4-7 hour half-life, allowing to normal 3-7 days after the stimulus is withdrawn. The ESR shows a slower rise and return to normal than C-reactive protein (CRP).

CRP versus ESR measurement

Erythrocyte sedimentation rate (ESR) is more commonly used as a non-specific marker of disease activity. However, as more is learned about CRP, measuring this parameter could be a better test than the ESR. The ESR, which is an indirect parameter of acute phase protein changes, can be influenced by concentrations of fibrinogen, monoclonal proteins and red cell morphology, whereas CRP has no cross-interfaces. CRP is useful for its negative predictive value as a negative CRP rules out the possibility of an inflammatory or necrotic course. A positive reaction is certainly an indication of a problem, but it is not specific for any single disease.

ESR has several disadvantages that prevent it from being an ideal laboratory test to monitor acute inflammation or tissue injury. However, the ESR remains useful for the detection of paraproteinaemia, which do not necessarily provoke an acute phase response. SLE and progressive systemic sclerosis, even when active, usually cause only a trivial increase in CRP (in the range 1-6 mg%), although the ESR may be very high .The reason for the discrepancy between ESR and CRP is unknown, but indicates the two tests are complementary. A comparison of ESR with CRP is shown in Table1.

Table 1 Comparison of CRP with Erythrocyte Sedimentation Rate

CONDITION OR VARIABLE	CRP	ESR
Specimen requirements	Serum or Plasma Stable in stored specimens	Fresh specimen of whole blood cannot be performed on stored specimen
Method of measurement	Direct quantitation of acute phase response	Indirect measurement of fibrinogen elevation
Magnitude and rate of rise	Elevation begins within 4 to 6 hrs, closely parallels acute response with 4 to 7 hrs. Half-life, allowing return to normal in 3 to 7 days after stimulus is withdrawn. Peak levels 100-1000 % above base line.	Rises more slowly, may not return to normal for weeks, despite clinical improvement. Fibrinogen increases up to 400% above base line.
Effects of anaemia, polycythemia, interaction of proteins and red blood cells, size, shape of red blood cells	Unaffected	False negative or false positive reactions, depending on abnormality.
Age and gender	Minimal change from neonate to elderly	Rises with age, higher values in women

LABORATORY METHODS OF MEASURING CRP

Latex Agglutination Assay

Traditional methods for measuring CRP include precipitation and agglutination assays. The latex agglutination assay is a qualitative test with a detection limit of approximately 10 mg/litre, the upper limit of normal. Because CRP levels can increase so rapidly and dramatically, the latex agglutination assay is subject to false-negative reactions due to a prozone-type phenomenon in which all of the antibody combining sites on the latex particles are bound to an excess of CRP so no cross-linking (agglutination) can occur. Consequently the qualitative tests should be performed on several dilutions of serum to avoid negative reactions. If several dilutions are formed, the latex agglutination method can easily be converted to a semi-quantitative assay so distinctions can be made between levels of positivity (e.g. less than 50 mg/litre and more than 150 mg/litre). Such semi-quantitative distinctions would be very useful to the clinician trying to distinguish between bacterial (high CRP levels) and viral infections (normal to slightly elevated CRP).

Immunoassays

Highly specific antibodies to CRP permit the development of rapid, specific, and very sensitive assays for this protein. These newer immunoassays include laser nephelometry (the most popular method), RIA, and enzyme immunoassays and have created a renewed interest in CRP testing in a variety of clinical settings. Measurement of CRP may be superior to the erythrocyte sedimentation rate (ESR) and may someday replace it. Recently, instrument manufacturers have developed assay systems that allow random access assays for

CRP to be performed virtually on demand with 10 to 20 minutes turn-around-time (TAT).

Ultra-sensitive or High-sensitivity (hs) CRP Assay

An ultra-sensitive immunoturbedimetric assay has been developed for CRP. The new assay measures the increased turbidity resulting from antibody-antigen complexes formed when sample and antibody reagent is mixed. The assay has sensitivity of 0.1 mg/litre. The ready-to-use liquid reagents can be placed directly on a chemistry analyser and will yield precise results in minutes (Cortlandt Manor, NY, USA).

Factors that affect results

As in all serological tests, haemolytic, lipemic or turbid sera may cause incorrect results and should not be used. Drugs that may cause false-positive results include oral contraceptives. Drugs that may cause false-negative results due to suppression of inflammation include NSAIDS, steroids and salicylates. The presence of intrauterine device may cause inflammation, which produces a positive test. Overnight refrigeration of the sample may produce a false-positive result. There is no need to refrigerate samples if the assay is to be performed on the same day. Demographic factors including age, sex and race should be used to adjust the upper reference limit for CRP.

STRUCTURE AND PHYLOGENY OF CRP

CRP belongs to the pentraxin family of calcium-dependent ligand-binding plasma proteins, the other member of which in

humans is serum amyloid P component (SAP). The human CRP molecule (M_r 115,135) is composed of five identical nonglycosylated polypeptide subunits (Mr 23,027), each containing 206 amino acid residues. The protomers are noncovalently associated in an annular configuration with cyclic pentameric symmetry (Figure 1).

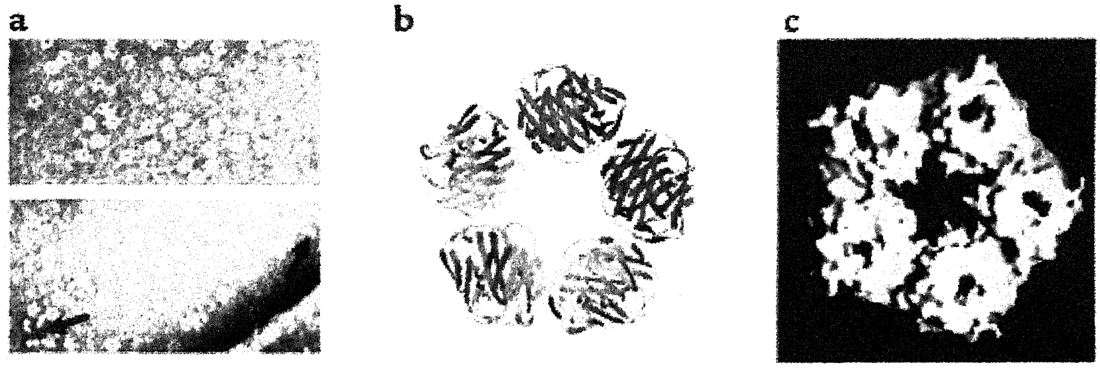


Figure 1.

Molecular structure and morphology of human CRP. (**a**) Negatively stained electron micrograph showing the typical pentameric disc-like structure face-on and side-on (arrows). (**b**) Ribbon diagram of the crystal structure, showing the lectin fold and the two calcium atoms (spheres) in the ligand-binding site of each protomer. (**c**) Space-filling model of the CRP molecule, showing a single phosphocholine molecule located in the ligand-binding site of each protomer.

Each protomer has the characteristic "lectin fold," composed of a two-layered β sheet with flattened jellyroll topology. The ligand-binding site, composed of loops with two calcium ions bound 4 Å apart by protein side-chains, is located on the concave face. The other face carries a single α helix (Figure 1). The pentraxin family, named for its electron micrographic appearance from the Greek *penta*

(five) *ragos* (berries), is highly conserved in evolution, with homologous proteins throughout the vertebrates and even in the phylo-genetically distant arachnid, *Limulus polyphemus*, the horseshoe crab. CRP function has largely been confined to passive administration of exogenous, heterologous CRP or to mice transgenic for rabbit or human CRP. These artifactual heterologous systems may not provide physiologically relevant information. Despite the evolutionary conservation of sequence, subunit organization, and protein fold, there are considerable variations between CRPs of different species with respect to fine ligand-binding specificity, presence and nature of glycosylation, protomer assembly, capacity to precipitate and aggregate ligands, base-line circulating concentrations, behavior as acute-phase proteins, and capacity to activate autologous complement. Indeed, only human CRP has been rigorously shown to activate complement in isologous serum. These differences command extreme caution in extrapolating from animal models to humans.

Human CRP binds with highest affinity to phosphocholine residues, but it also binds to a variety of other autologous and extrinsic ligands, and it aggregates or precipitates the cellular, particulate, or molecular structures bearing these ligands. Autologous ligands include native and modified plasma lipoproteins, damaged cell membranes, a number of different phospholipids and related compounds, small nuclear ribonucleoprotein particles, and apoptotic cells. Extrinsic ligands include many glycan, phospholipid, and other constituents of microorganisms, such as capsular and somatic components of bacteria, fungi, and parasites, as well as plant products. When aggregated or bound to macromolecular ligands,

human CRP is recognized by C1q and potently activates the classical complement pathway, engaging C3, the main adhesion molecule of the complement system, and the terminal membrane attack complex, C5–C9. Bound CRP may also provide secondary binding sites for factor H and thereby regulate alternative-pathway amplification and C5 convertases.

The secondary effects of CRP that follow ligand binding resemble some of the key properties of antibodies, suggesting that under various circumstances CRP may contribute to host defense against infection, function as a proinflammatory mediator, and participate in physiological and pathophysiological handling of autologous constituents. Evidence of CRP functioning in these various roles is available from experimental animal models, but there is no rigorous information from physiological isologous systems. The absence of any known deficiency or protein polymorphism of human CRP, and the phylogenetic conservation of CRP structure and its ligand-binding specificity for phosphocholine and related substances, suggest that this protein must have had survival value. Microbial infection is a major driving force of change during evolution, and CRP has many features compatible with a role in innate immunity. In addition, the impaired CRP response in active systemic lupus and the marked spontaneous antinuclear autoimmunity of SAP knockout mice are compatible with the possibility that pentraxins function to prevent autoimmunity.

Phosphocholine is a component of many prokaryotes and is almost universally present in eukaryotes, and a substantial proportion of germline-encoded, highly conserved natural antibodies resemble

CRP in specifically recognizing phosphocholine. The capacity to bind these residues may thus be important for both host defense and handling of autologous constituents including necrotic and apoptotic cells. Activation of complement by human CRP may then opsonize and enhance phagocytosis of these various ligands but could also mediate proinflammatory pathophysiological effects. Intriguingly, the spectrum of autologous ligands recognized by CRP overlaps that of anti-phospholipid autoantibodies that are associated with premature cardiovascular disease in autoimmune syndromes.

Some functions that have been claimed for CRP seem inherently unlikely. For example, it is improbable that a plasma protein with a dynamic range of 10,000-fold within hours would function like a cytokine or be a fine modulator of sophisticated cellular or physiological systems. Another implausible speculation concerns dissociated denatured CRP subunits, so-called modified or neo-CRP, for which various biological effects has been reported in vitro. Native CRP is actually very stable, and release of separate protomers requires exposure of the protein to harsh denaturing conditions. There is no compelling evidence for the persistence of denatured CRP in vivo, and rapid complete catabolism of such material would be expected.

A large number of changes, distant from the site or sites of inflammation and involving many organ systems, may accompany inflammation. In 1930 interest was focused on these changes by the discovery of C-reactive protein (so named because it reacted with the pneumococcal C-polysaccharide) in the plasma of patients during the acute phase of pneumococcal pneumonia. Accordingly, these

systemic changes have since been referred to as the acute-phase response, even though they accompany both acute and chronic inflammatory disorders.

Acute Phase Response

Inflammation is a protective reaction of vascular connective tissue to damaging stimuli, including infection. The inflammatory response is associated with vasodilatation, increased vascular permeability, recruitment of inflammatory cells (especially neutrophils in acute inflammation), the release of inflammatory mediators from these cells (including vasoactive amines, prostanoids, and reactive oxygen intermediates), and cytokine release. The macrophage-derived cytokines IL-1 and IL-6 are primarily responsible for the acute phase response, a protective change in plasma protein production by hepatocytes.

Table 2: Acute Phase Proteins Increased

Protease Inhibitors	□$_1$-antitrypsin antichymotrypsin	Inter□-antitrypsin
Coagulation Proteins	Fibrinogen Prothrombin Factor VIII Plasminogen	
Complement Proteins	C1s, C2, C3, C4, C5 Factor B C1 esterase inhibitor Plasminogen	Properdin
Transport Proteins	Haptoglobin Haemopexin Caeruloplasmin	
Miscellaneous	**C-reactive protein** Serum amyloid A protein Fibronectin $_1$-acid glycoprotein	Albumin Pre-albumin High- and low-density lipoprotein

Acute-phase changes may be divided into changes in the concentrations of many plasma proteins, known as the acute-phase proteins (Table 2), and a large number of behavioral, physiologic, biochemical, and nutritional changes. An acute-phase protein has been defined as one whose plasma concentration increases (positive acute-phase proteins) or decreases (negative acute-phase proteins) by at least 25 percent during inflammatory disorders. The changes in the concentrations of acute-phase proteins are due largely to changes in

their production by hepatocytes. The magnitude of the increases varies from about 50 percent in the case of ceruloplasmin and several complement components to as much as 1000-fold in the case of C-reactive protein and serum amyloid A.

Conditions that commonly lead to substantial changes in the plasma concentrations of acute-phase proteins include infection, trauma, surgery, burns, tissue infarction, various immunologically mediated and crystal-induced inflammatory conditions, and advanced cancer. Moderate changes occur after strenuous exercise, heatstroke, and childbirth. Small changes occur after psychological stress and in several psychiatric illnesses. Although the concentrations of multiple components of the acute-phase response commonly increase together, not all of them increase uniformly in all patients with the same illness. Thus, febrile patients may have normal plasma concentrations of C-reactive protein, and discordance between the plasma concentrations of different acute-phase proteins is common. These variations, which indicate that the components of the acute-phase response are individually regulated, may be explained in part by differences in the patterns of production of specific cytokines or their modulators in different pathophysiologic states.

Induction of Acute-Phase Proteins by Cytokines and Other Extracellular Signaling Molecules Cytokines are intercellular signaling polypeptides produced by activated cells. Most cytokines have multiple sources, multiple targets, and multiple functions. The cytokines that are produced during and participate in inflammatory processes are the chief stimulators of the production of acute-phase proteins. These inflammation-associated cytokines include

interleukin-6, interleukin-1ß, tumor necrosis factor α, interferon-γ, transforming growth factor ß, and possibly interleukin-8. They are produced by a variety of cell types, but the most important sources are macrophages and monocytes at inflammatory sites.

Interleukin-6 is the chief stimulator of the production of most acute-phase proteins, whereas the other implicated cytokines influence subgroups of acute-phase proteins. However, in mice rendered incapable of expressing interleukin-6 (knockout mice), the role of interleukin-6 in stimulating the production of acute-phase proteins depends on the nature or site of the inflammatory stimulus; the response is largely inhibited in interleukin-6 knockout mice injected with turpentine but is normal when bacterial lipopolysaccharide is the inflammatory stimulus. This finding indicates that lipopolysaccharide causes the production of other cytokines capable of stimulating the production of acute-phase proteins. The responses are similar in interleukin-1ß knockout mice, presumably because interleukin-1ß is required to stimulate the production of interleukin-6 after the administration of turpentine. These studies provide further evidence that patterns of cytokine production and the acute-phase response differ in different inflammatory conditions. Interleukin-11, leukemia inhibitory factor, oncostatin M, ciliary neurotrophic factor, and cardiotrophin 1 may have actions similar to those of interleukin-6.

Cytokines operate both as a cascade and as a network in stimulating the production of acute-phase proteins. Many cytokines can regulate the production of other cytokines and cytokine receptors. For example, tumor necrosis factor αis the main stimulator

of interleukin-1 production in patients with rheumatoid arthritis; interleukin-1ß may increase or decrease expression of its own receptors; the interleukin-6 response to the injection of turpentine in mice requires interleukin-1ß; and interleukin-6 inhibits the expression of tumor necrosis factor α.

In addition, cytokines are components of a large, complex signaling network. Most likely, cells are seldom exposed to only a single cytokine. Instead, combinations of mediators convey biologically relevant information. The effects of cytokines on target cells may be inhibited or enhanced by other cytokines, by hormones, and by cytokine-receptor antagonists and circulating receptors. Combinations of cytokines have been found to have additive, inhibitory, or synergistic effects. Thus, the induction of C-reactive protein and serum amyloid A in some models requires both interleukin-6 and either interleukin-1 or tumor necrosis factor α, and the induction of fibrinogen by interleukin-6 is inhibited by interleukin-1, tumor necrosis factor α, and transforming growth factor ß. Interleukin-6 enhances the effect of interleukin-1ß in inducing the expression of interleukin-1–receptor antagonist, and interleukin-4 inhibits the induction of some acute-phase proteins by other cytokines. Soluble interleukin-6 receptor αmolecules increase the effects of the ligand, whereas other soluble receptors, such as those for tumor necrosis factor $\alpha$and interleukin-1, are inhibitory. Glucocorticoids generally enhance the stimulatory effects of cytokines on the production of acute-phase proteins, whereas insulin decreases their effects on the production of some acute-phase proteins.

The expression of genes for acute-phase proteins is regulated mainly at the transcriptional level, but post-transcriptional mechanisms also participate. Post-translational changes in the glycosylation of plasma proteins during inflammatory states include alterations in oligosaccharide branching, increased sialylation of orosomucoid, and decreased galactosylation of IgG. Changes in oligosaccharide branching are induced by inflammation-associated cytokines, independently of their effects on the production of acute-phase proteins. Finally, the efficiency of secretion of C-reactive protein, a process distinct from its synthesis, is greatly increased during the acute-phase response.

Postulated Function of the Acute-Phase Response

The assumption that the changes in plasma concentrations of acute-phase proteins are beneficial is based largely on the known functional capabilities of the proteins and on logical speculation as to how these might serve useful purposes in inflammation, healing, or adaptation to a noxious stimulus. Inflammation is a complex, highly orchestrated process involving many cell types and molecules, some of which initiate, amplify, or sustain the process, some of which attenuate it, and some of which cause it to resolve. A number of the participating molecules are multifunctional and contribute to both the waxing and the waning of inflammation at different points in its evolution.

Many of the acute-phase proteins have the potential to influence one or more of these stages of inflammation. A major function of C-reactive protein, a component of the innate immune system, is its ability to bind phosphocholine and thus recognize some

foreign pathogens as well as phospholipid constituents of damaged cells. It can activate the complement system when bound to one of its ligands and can also bind to phagocytic cells, an observation suggesting that it can initiate the elimination of targeted cells by its interaction with both humoral and cellular effector systems of inflammation. Other proinflammatory effects of C-reactive protein include the induction of inflammatory cytokines and tissue factor in monocytes. However, its net effect is antiinflammatory in transgenic mice that produce large amounts of C-reactive protein. Such an effect of C-reactive protein may be explained by its ability to prevent the adhesion of neutrophils to endothelial cells by decreasing the surface expression of L-selectin, to inhibit the generation of superoxide by neutrophils, and to stimulate the synthesis of interleukin-1–receptor antagonist by mononuclear cells. It seems likely that C-reactive protein has many pathophysiologic roles in the inflammatory process.

Several acute-phase proteins initiate or sustain inflammation. The classic complement components, many of which are acute-phase proteins, have central proinflammatory roles in immunity, as does mannose-binding lectin, a recently recognized acute-phase component of complement. Complement activation leads to chemotaxis, plasma protein exudation at inflammatory sites, and opsonization of infectious agents and damaged cells. Similarly, granulocyte colony-stimulating factor increases the inflammatory response by increasing the numbers of granulocyte precursors in bone marrow and by activating mature granulocytes.

Clinical Assessment of Acute-Phase Proteins and Cytokines

Estimation of other changes in acute-phase proteins, despite the lack of diagnostic specificity, is useful to clinicians because such changes reflect the presence and intensity of an inflammatory process. Thus, measurements of plasma or serum C-reactive protein can help differentiate inflammatory from noninflammatory conditions and are useful in managing the patient's disease, since the concentration often reflects the response to and need for therapeutic intervention. Finally, in some diseases, such as rheumatoid arthritis, serial measurements of C-reactive protein are of prognostic value. In evaluating laboratory results, physicians should be aware that some laboratories report C-reactive protein concentrations in milligrams per liter and others in milligrams per deciliter.

Currently, the most widely used indicators of the response of acute-phase proteins are the erythrocyte sedimentation rate and the plasma C-reactive protein concentration. The rate at which erythrocytes fall through plasma — that is, the erythrocyte sedimentation rate — depends largely on the plasma concentration of fibrinogen. As a test, the erythrocyte sedimentation rate has the advantages of familiarity, simplicity, and an abundant literature compiled over the past seven decades. Nonetheless, measurement of C-reactive protein has several advantages over this method. The erythrocyte sedimentation rate is an indirect measurement of plasma acute-phase protein concentrations and can be greatly influenced by the size, shape, and number of erythrocytes, as well as by other plasma constituents such as immunoglobulins. Consequently, the results are imprecise and sometimes misleading. Although the

erythrocyte sedimentation rate represented a great advance when it was introduced in the 1920s, this indirect method (which some regard as archaic) is no longer needed to assess plasma concentrations of fibrinogen, because they can now be determined directly. As a patient's condition worsens or improves, the erythrocyte sedimentation rate changes relatively slowly, whereas plasma C-reactive protein concentrations change rapidly. The range of abnormal values for C-reactive protein is broader than the range of abnormal values for the erythrocyte sedimentation rate, with accompanying clinical implications: among patients with plasma C-reactive protein concentrations greater than 100 mg per liter, 80 to 85 percent have bacterial infections. The erythrocyte sedimentation rate increases steadily with age, but plasma C-reactive protein concentrations do not.

Conclusions

The acute-phase response, an important pathophysiologic phenomenon, replaces the normal homeostatic mechanisms with new set points that presumably contribute to defensive or adaptive capabilities. The functions of these changes are highly variable and diverse: some participate in initiating or sustaining the inflammatory process, others modulate it, and still others have adaptive roles. These changes are induced by a complex intercellular signaling system of which the chief constituents are inflammation-associated cytokines. Several cytokines, particularly interleukin-6, stimulate the production of acute-phase proteins in response to varied stimuli. The patterns of cytokine production and of the acute-phase response differ in different inflammatory conditions. Acute-phase changes

reflect the presence and intensity of inflammation, and they have long been used as a clinical guide to diagnosis and management. For this purpose, determination of serum C-reactive protein has advantages over the traditional strategy of measuring the erythrocyte sedimentation rate.

CRP levels can increase to as much as 1–1000 fold from baseline concentrations with bacterial infection, trauma, surgery, and other inflammatory events, declining to baseline level in 12–14 days.

In healthy young adult volunteer blood donors, the median concentration of CRP is 0.8 mg/l, the 90th centile is 3.0 mg/l, and the 99th centile is 10 mg/l, but, following an acute-phase stimulus, values may increase from less than 50 μg/l to more than 500 mg/l, that is, 10,000-fold. Plasma CRP is produced only by hepatocytes, predominantly under transcriptional control by the cytokine IL-6, although other sites of local CRP synthesis and possibly secretion have been suggested. De novo hepatic synthesis starts very rapidly after a single stimulus, serum concentrations rising above 5 mg/l by about 6 hours and peaking around 48 hours. The plasma half-life of CRP is about 19 hours and is constant under all conditions of health and disease, so that the sole determinant of circulating CRP concentration is the synthesis rate, which thus directly reflects the intensity of the pathological process(es) stimulating CRP production. When the stimulus for increased production completely ceases, the circulating CRP concentration falls rapidly, at almost the rate of plasma CRP clearance. In most, though not all, diseases, the circulating value of CRP reflects ongoing inflammation and/or tissue damage much more accurately than do other laboratory parameters of

the acute-phase response, such as plasma viscosity and the erythrocyte sedimentation rate. Importantly, acute-phase CRP values show no diurnal variation and are unaffected by eating. Liver failure impairs CRP production, but no other intercurrent pathologies and very few drugs reduce CRP values unless they also affect the underlying pathology providing the acute-phase stimulus. The CRP concentration is thus a very useful nonspecific biochemical marker of inflammation, measurement of which contributes importantly to (a) screening for organic disease, (b) monitoring of the response to treatment of inflammation and infection, and (c) detection of intercurrent infection in immunocompromised individuals, and in the few specific diseases characterized by modest or absent acute-phase responses.

Since CRP is itself an acute phase reactant, its measure reflects the level of inflammation directly, rather than the more indirect reflection of inflammation that the ESR provides. CRP production is induced by the liver under the influence of IL-1 and IL-6. It reaches peak levels quickly, approximately 50 hours and also falls rapidly once the stimulus is removed. Thus the CRP provides a more immediate picture of the level of inflammation than does the ESR in which fibrinogen level rises and falls more slowly. Its function in-vivo is felt to be to assist in the activation of the complement system, influence phagocytic cell function, and augment cell mediated cytotoxicity i.e. amplify the immune response.

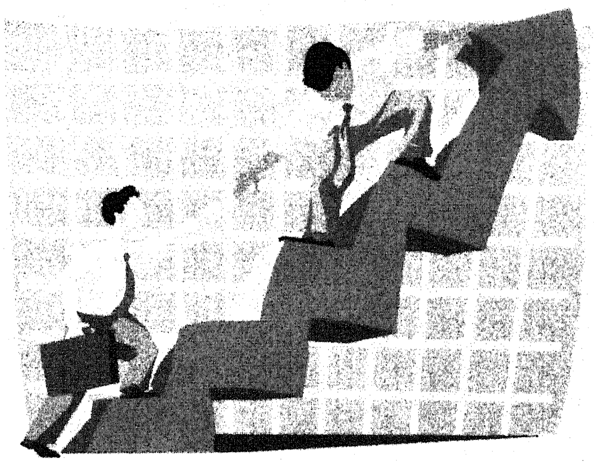
Evans MI & et al (1980) found that elevated C-reactive protein very accurately divided patients with evidence of infectious morbidity from those without such evidence. C-reactive protein

elevated at least 12 hours prior to any other parameter measured. They suggested that C-reactive protein may be reliable, early predictor of infectious morbidity and thus may be of benefit in the selective management of patients of premature rupture of membranes.

Elevated C-reactive protein levels correlated well with both the pathologic and clinical diagnosis of chorioamnionitis. Elevated C-reactive protein levels are more sensitive than other standard laboratory or clinical tests in predicting chorio-amnionitis both by clinical and pathological criteria (Ismail MA, 1985).

Determination of serum CRP can also be used to help to confirm or rule out bacterial infections in the neonatal period, for even premature babies have the capacity to synthesise CRP in the liver if they contract an infection.

AIMS & OBJECTIVES



AIMS & OBJECTIVES

The present clinical study was undertaken to evaluate the following aims and objectives:

1. Early detection of infection for considering conservative management of patients with PROM.

2. To diagnose chorioamnionitis before clinical symptoms appear.

3. To evaluate the use of C-reactive protein as a possible marker for subclinical chorioamnionitis.

4. Early diagnosis of neonatal infection.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Tillet et al (1930) observed that sera obtained from patient during febrile illnesses precipitated with c-polysaccharides, extract of pneumococcus first designated fraction C.

Fraction C substance was subsequently proved to be a protein designated as C-reactive protein by Abernathy and Aveny in 1941.

CRP, named for its capacity to precipitate the somatic C-polysaccharide of *Streptococcus pneumoniae*, was the first acute-phase protein to be described and is an exquisitely sensitive systemic marker of inflammation and tissue damage. The acute-phase response comprises the nonspecific physiological and biochemical responses of endothermic animals to most forms of tissue damage, infection, inflammation, and malignant neoplasia. In particular, the synthesis of a number of proteins is rapidly upregulated, principally in hepatocytes, under the control of cytokines originating at the site of pathology.

CRP belongs to the pentraxin family of calcium-dependent ligand-binding plasma proteins. The intact CRP molecule is a pentameric protein with identical subunits arranged in a doughnut-shaped polymer. The human CRP molecule (M_r 115, 135) is composed of five identical nonglycosylated polypeptide subunits (Mr 23,027), each containing 206 amino acid residues. The protomers are noncovalently associated in an annular configuration with cyclic pentameric symmetry. Each protomer has the characteristic "lectin

fold," composed of a two-layered β sheet with flattened jellyroll topology. The ligand-binding site, composed of loops with two calcium ions bound 4 Å apart by protein side-chains, is located on the concave face. The other face carries a single α helix. The pentraxin family, named for its electron micrographic appearance from the Greek *penta* (five) *ragos* (berries), is highly conserved in evolution, with homologous proteins throughout the vertebrates and even in the phylo-genetically distant arachnid, *Limulus polyphemus*, the horseshoe crab.

Harlimann et al (1966) found that C-reactive protein is exclusively synthesized in liver.

Ramos and Stern (1969) were of the opinion that examination of gastric aspirate of cells while indicating numerically greater number of suspected than subsequently proved infection, nevertheless be used as a rapid, simple and effective mean of identifying the infant at risk.

Gunn et al (1970) in a study at the university of California in Los Angeles found that incidence of overall premature rupture of membrane ranging from 2-18% and they found that labour started within 24 hours of premature rupture of membrane in 81% of patients carrying babies larger than 2.5 Kg. the prevalence of chorioamnionitis was 2.7% before 12 hours, 6.3% between 12 & 24 hours and 26.4% after 24 hours of latent period between rupture of membranes and delivery of baby.

Bobitt et al and hosmer et al (1972) shown a direct co-relation between duration of leaking and frequency of early neonatal infection.

Canlon J (1972) suggested that gastric aspirate cellularity might be used to screen out the newborn that are likely to develop infection.

Kusner and his associates (1973) identified maternal fever during labour, premature rupture of membranes, asphyxia and few other problems not resulting from infections to be associated with elevated level of CRP in umbilical cord blood.

Takkar V.P. et al (1973) found that incidence of early neonatal infection was higher in cases where amniotic membrane had ruptured for more than 24 hours, before delivery as compared to shorter duration of rupture of membrane and incidence of early neonatal infection has been reported to vary from 3% to 13.2% when duration of rupture of membranes exceeds 24 hours and increases the risk of septicemia.

Sable and Hanson (1974) observed high C-reactive protein values during first few hours after clinical symptoms had appeared. This suggested that C-reactive protein was sufficiently rapid and specific to serve as a definite aid in early diagnosis of septicemia.

According to Artae et al (1976) poor nutritional status, which is significantly influenced, by the socioeconomic status of the patients is associated with premature rupture of membranes.

Claus et al (1976) observed that CRP in neonate is endogenously produced.

Khudsen et al (1976) mentioned that the risk of septicemia is highest in the infants of low birth weight and in presence of neonatal asphyxia.

The occurrence of chorioamnionitis infection after premature rupture of membranes seems to be greater in hospitals caring for low socioeconomic segments of the population than in institution taking care of the affluent. The reason for this difference is decreased antibacterial activity in the amniotic fluid of low socioeconomic group (Tagari et al, 1977).

Sable and Wadsworth (1979) using latex agglutination test found that in 94% of non infected infants had C-reactive protein 15mg/litre and 82% had C-reactive protein 10 mg/litre upto 3 days of age. After 3 days of age 96% had C-reactive protein 10mg/litre. Level of C-reactive protein rises to a far greater magnitude in bacterial illness as against viral illness (Pepys MB 1981) and not altered by drugs as steroids.

Matesanz JL, Malaga S, Santos F, Nuno F, Ramos A, Crespo M. & associates (1979) determined serum levels of C-reactive protein (C.R.P.) in a series of 125 children under one month age. Of them, 75 were clinically healthy and 50 showed signs that suggested infection. In all this infants, in addition to C.R.P., different components of white blood cell differential count were determined, and a bacteriological study was undertaken. They found high concentration

of C.R.P. in 100% of children in which diagnosis of sepsis was bacteriologically confirmed. On the other hand, concentrations of this biological parameter were not substantially modified in newborns used in the control, nor in those other ones in which sepsis was not confirmed. Sensibility of C.R.P. showed itself significantly higher than other hematological indexes in study.

In 1980 **Evans MI, Hajj SN, Devoe LD, Angerman NS, Moawad AH.** and associates found that the management of patients with premature rupture of membranes poses one of the most serious dilemmas since it significantly increases the likelihood of prematurity and serious perinatal infection. Early infection is not reliably predicted nor detected by standard laboratory parameters. Serum C-reactive protein levels were assayed along with WBC count, DLC count and temperature course in patients with premature rupture of membranes. Elevated C-reactive protein very accurately divided patients with evidence of infectious morbidity from those without such evidence ($p < 0.001$) .In 109 patients there were 11 false negative and no false positive. In 14 of 20 patients followed with serial comparison who developed morbidity, C-reactive protein elevated at least 12 hours prior to any other parameter measured. Changes in the other six patients were concurrent. The results suggests that C-reactive protein may be reliable, early predictor of infectious morbidity and thus may be of benefit in the selective management of patients of premature rupture of membranes.

According to Naey and Peters (1980) risk factors for premature rupture of membranes are advanced maternal age, non-white race, multiparity, instrumentation of cervix prior to pregnancy, cigarette

smoking, incompetent cervix, low pregnancy weight gain recent coitus.

In 1981 John WC, Johnson, Norman H, Daikoku Jannifer, R. Niebyl and associates conducted a retrospective study in 8321 patients with premature rupture of membranes to determine the consequences of prolongation of latent period. Among patients with pregnancies of more than 37 weeks duration, those with premature rupture of membranes and latent periods of more than one day demonstrated an increased incidence of intra-partum fever; whereas those with latent periods of more than three days demonstrated a marked increased in foetal (but not neonatal) deaths.

Although intrapartum fever and perinatal mortality were more common in preterm pregnancies, neither was found to increase or decrease with prolonged latency, provided differences in gestational ages and race were taken into account .In the absence of chorio-amnionitis, there appears to be no benefit of delivery before 37 weeks gestation.

Ainbender E, Cabatu EE, Guzman DM, Sweet AY. et al (1982) found that infant as immature as 28 weeks gestation were able to produce C-reactive protein concentration 2 mg/dl. He added shock, foetal distress and aspiration difficulties to the conditions in which serum C-reactive protein values are commonly high at birth and during the first few days of life and eliminated C-reactive protein as a useful indicator of infection.

Harry F. Frab, Mark Arnesen and associates (1983) determined C-reactive protein serially in 31 patients with premature rupture of membranes, 41 patients in premature labour and 18 pregnant patients with a variety of high risk condition. Elevated levels of C-reactive protein were not predictive of clinical amnionitis, histological chorioamnio-nitis, or neonatal sepsis. No discernible relationship was found between serum C-reactive protein and peripheral white blood cell count.

C-reactive protein was not elevated (false negative) in two patients in premature labour group with proved bacterial amnionitis. Elevated C-reactive protein in the absence of infection (false positive) likewise occurred. The results suggest that C-reactive protein can be used in conjunction with other signs and symptoms suggestive of chorioamnionitis rather than as a pathognomic test.

In 1983 **Hawrylyshyn P, Bernstein P, Milligan JE, Soldin S, Pollard A, Papsin FR.** evaluated a group of 52 patients with premature rupture of membranes before 34 weeks gestation prospectively and managed expectantly. Of 42 patients who were delivered of their infants, 26 (61.9%) had significant chorioamnionitis on histopathology, and 18 had positive microbial cultures at delivery. However, only seven patients (16.7%) developed clinical signs of chorioamnionitis. There were no maternal deaths or perinatal deaths attributed to sepsis. Only two infants (< 5%) had positive blood culture. All patients were assessed daily for the development of chorioamnionitis. Amniocentesis was not routinely performed. White blood cell counts, band neutrophile count, and erythrocyte sedimentation rate were found to be unreliable. C-

reactive protein determination was found most reliable with a high degree of sensitivity and specificity. Elevated C-reactive protein levels correlated better with pathologic confirmation of chorioamnionitis than with the clinical febrile morbidity.

Kumari and Kayal et al (1983) found that septicemia originating as pulmonary focus kills the neonates in most cases. Respiratory distress in the newborn can be due to congenital pneumonia and they found that gastric aspirate polymorphs a useful side laboratory diagnostic test, the incidence of septicemia bearing a direct association to the number of gastric aspirate polymorphs / high power field.

According to Lnihan (1984) antenatal vaginal examination is associated with premature rupture of membranes.

Handwerker SM, Tejani NA, Verma UL, Archbald F. (1984) & associates found that Subclinical intrauterine infection was an important cause of preterm labor, specifically where tocolysis had failed. Fifty patients in preterm labor with singleton pregnancies were studied prospectively to determine whether the presence or absence of C-reactive protein, a nonspecific marker for infection, would correlate with success or failure of tocolysis. Of the 50 patients, tocolysis failed in 11 of 15 women with a positive C-reactive protein determination. Tocolysis succeeded in 33 of 35 cases where C-reactive protein was negative (P less than .0005). Urinary tract infection occurred in 40% of the study patients, but was not a confounding factor in the interpretation of C-reactive protein.

Romem Y, Artal R. (1984) evaluated C-reactive protein for its ability to predict the occurrence of clinical chorioamnionitis in 51 patients with spontaneous premature rupture of the membranes at less than or equal to 34 weeks of gestational age. All the patients had determinations of C-reactive protein on admission, and then 25 patients were tested daily. Of the total 51 patients, 14 developed clinical signs compatible with a diagnosis of chorioamnionitis. An analysis was conducted to compare the use of C-reactive protein to that of white blood cell count in predicting febrile disease. Their study indicates that C-reactive protein is an accurate and early marker for predicting clinical chorioamnionitis. White blood cell and differential counts are less accurate in such prediction, especially after steroid treatment.

Andrade Vargas A, del Carmen Leon Ramirez N, Santa Maria Favela D, Oseguera Villanueva R. (1984) in a prospective study showed prognostic value of C-reactive protein in premature rupture of membranes.

In 1984 **O'Callaghan C, Franklin P, Elliott TS, Deverill I, Richards N, Powell RJ.** used a latex enhanced immunoassay on a centrifugal fast analyser to compare serum C reactive protein concentrations in maternal and neonatal blood. In the neonate the C reactive protein concentration at birth was less than 1.0 mg/l; the concentration rose slightly during the first two weeks of life. There was no correlation between C reactive protein concentrations in maternal and neonatal sera. No significant difference was found between the C reactive protein concentrations in blood obtained by either heel prick or venepuncture.

Philip AG. (1984) Showed no single diagnostic test for neonatal sepsis is both rapid and reliable. Combining leukocyte (WBC) counts with acute phase reactants (APR) enhances diagnostic accuracy. The most helpful WBC counts are leukopenia (less than 5.0 x 10(9)/l), increased immature/total neutrophils (greater than or equal to 0.2) and profound neutropenia (less than 1.0 x 10(9)). Of the APR, C-reactive protein responds most rapidly, but alpha 1-acid glycoprotein (orosomucoid), haptoglobin and mini-ESR (greater than or equal to 15 mm/h) are also useful. Rapid, quantitative determinations of APR are now available with nephelometric techniques. Abnormal WBC counts frequently appear before APR changes in group B streptococcal infection. Sequential determinations of WBC counts and APR may provide valuable diagnostic and prognostic information.

Hindocha P, Campbell CA, Gould JD, Wojciechowski A, Wood CB. (1984)& Associates assayed serial C-reactive protein concentrations by electro immunoassay in 41 infants. Values in most of the non-infected infants were below 0.3 mg/dl, the lower limit of detection of C-reactive protein by electro immunoassay. Eleven of 12 infants with proved sepsis (positive blood cultures) had significantly raised concentrations and one infant with recurrent pseudomonas chest infection had a raised C-reactive protein concentration. High C reactive protein concentrations were also found in infants with suspected infection. Successful treatment was followed by a decrease in the C reactive protein concentration. Total white blood cell count was not as appropriate as C-reactive protein determination in the early identification of bacterial infection in the newborn.

Hameed C, Tejani N, Verma UL, Archbald F. in (1984) evaluated thirty-seven consecutive patients with singleton pregnancies in "uncomplicated" preterm labor with intact membranes suitable for tocolysis for evidence of silent chorioamnionitis by means of maternal serum C-reactive protein and amniotic fluid white blood cell count, Gram stain, and cultures. Abnormalities in these markers of infection were found to be significantly more common in cases that were refractory to tocolysis. These cases also showed both pathologic evidence of chorioamnionitis and a significantly greater neonatal early infectious morbidity. They concluded that silent chorioamnionitis is a significant cause of "uncomplicated" preterm labor refractory to conventional methods of tocolysis.

Seo K, McGregor JA, French JI. Conducted a retrospective study of 9642 births at the University of Colorado Health Sciences Center between July 1980 and June 1985, to evaluate possible associations between preterm birth and maternal and neonatal infections. Clinical chorioamnionitis occurred more frequently among women delivering before term with intact membranes at the onset of labor (5.8% preterm versus 1.7% term) and among women with PROM (26.5% preterm versus 6.7% term). Among the women delivered by cesarean, the incidence of postpartum endometritis was higher in those with preterm PROM than in those with term rupture of membranes. The incidence of neonatal infection increased significantly as the gestational age of the neonates decreased (P less than .01). The rate of culture-proven neonatal infection was significantly higher following PROM (P less than .01) than after birth without PROM. Both neonatal infection and perinatal mortality were increased in association with chorioamnionitis in both preterm

and term pregnancies. These consistent observations complement and support suggestions that reproductive tract infection plays a possibly preventable role in the pathogenesis of preterm birth.

Reports of Polansky GH et al (1985) showed the incidence PROM ranges from 14-17%. Overall, about 10% of all gestations are complicated by PROM. At term, the incidence of PROM varies from 6 to 19%.

Kite P, Millar MR, Gorham P, Congdon P. (1985) evaluated the neutrophil count, immature:total neutrophil ratio, C reactive protein assay, nitroblue tetrazolium test and an acridine orange leucocyte cytospin test for the diagnosis of neonatal bacteraemia. The acridine orange leucocyte cytospin test gave the highest specificity and positive predictive accuracy, but was less sensitive than the neutrophil count, C reactive protein assay or nitroblue tetrazolium test, particularly for the diagnosis of bacteraemia caused by coagulase negative staphylococci. No single test gave the sensitivity, specificity, and positive predictive accuracy of the combined results of the acridine orange leucocyte cytospin, C reactive protein, and nitroblue tetrazolium tests

Levels of C-reactive protein rise within hours to significant levels after inflammatory reaction **(Malaviya A.N., Singh G., 1985).**

In 1985 **Ismail MA, Zinaman MJ, Lowensohn RI, Moawad AH**. And associates conducted a retrospective study of 100 patients with premature of membranes, clinical chorioamnionitis was present in 18 cases and histological chorioamnionitis was present in 65

cases. Patients who were managed conservatively for premature rupture of membranes were monitored by C-reactive protein determination, white blood cell count, and differential counts, maternal temperature and foetal heart rate. Elevated C-reactive protein levels correlated well with both the pathologic and clinical diagnosis of chorioamnionitis. Elevated C-reactive protein levels (at least 1 to 24 hours before delivery) were more sensitive than other standard laboratory or clinical tests in predicting chorio-amnionitis both by clinical and pathological criteria. When C-reactive protein values were normal, clinical chorioamnionitis was rarely found, whereas pathologically diagnosed chorioamnionitis was found half of the time. Thus they concluded that although the C- reactive protein level is a very sensitive predictor of infectious morbidity in premature rupture of membranes but its specificity is not high.

Adhikari M, Coovadia HM, Coovadia YM, Smit SY, Moosa A. (1986) studied the following non-specific indices of infection in septicaemic and non-septicaemic babies: haemoglobin, total white blood cell count, differential white count, ESR, platelet count, C-reactive protein (CRP), serum immunoglobulins, plasma C3 and haptoglobin. Forty-three low-birthweight (LBW) infants with clinical features suggesting septicaemia were investigated; blood cultures were positive in 19 and negative in 24. The mortality was 53% in the culture-positive and 13% in the culture negative group. Comparisons between the two groups of babies showed that the CRP titre (measured by Latex agglutination) was the only reliable non-specific indicator of infection. The titre was elevated more often in culture positive (16/19) than culture negative (7/17) babies (P less than 0.001). The CRP titre (Mean +/- 2 S.D.) was 15.75 +/- 12 in blood

culture positive and 6.13 +/- 11.72 in culture negative neonates respectively (P less than 0.0004). Positive CRP titres were found in 5 of 20 healthy controls (4 +/- 8.4). Sequential CRP titres showed a gradual decline with clinical improvement in both groups of patients. The IgM was unhelpful as it was raised (greater than or equal to 40 mg%) in 37 of the patients.

In 1987 **Nicholas M. Fisk, John Fish, Andrew G, Child and associates** conducted in a prospective blind study 380 daily serum samples from 55 women with preterm pre-mature rupture of membranes were analysed for C-reactive protein. Although the last C-reactive protein before delivery was higher in patients with histological chorioamnionitis (p<.007), considerable overlap between infected and non-infected pregnancies occurred. Precluding the use of C-reactive protein as diagnostic test if published normal levels were used. When upper limits were set at 30, 35, or 40 mg/liter; the last C-reactive protein before delivery proved 90, 95 and 100% positively predictive of infection in singleton pregnancies. Such high specificities are needed to prevent inappropriate intervention based on false positive results. They therefore proposes upper limits for single estimation of 30, 35, or 40 mg/liter depending on the relative risk of prater delivery versus infection at various gestational ages. In addition, consecutive value 20 mg/l appeared highly predictive of infection.

Morales WJ (1987) conducted a prospective study in 698 women between 26-34 weeks with preterm premature rupture of membranes and reported that 13% mothers developed chorioamnionitis and infants born to mothers with chorioamnionitis

had a four fold increased incidence of neonatal mortality and three fold increased incidence of respiratory distress syndrome, neonatal sepsis and intraventricular haemorrage.

Salzen HR, Genger H, Muhar U, Lischka A and his associates (1987) determined the C-reactive protein concentration in 25 infants whose mothers had presented with pro-longed rupture of amniotic membranes, premature rupture of membranes and / or amnionitis. C-reactive protein was positive (i.e. greater than or equal to 6 mg/l) within first 6 hours of life in 10 and negative in15 infants. Clinically all infants with positive C-reactive developed symptoms suggestive of bacterial infection and both the absolute immature neutrophil counts as well as the ratio immature /total neutrophil were significantly higher in them on day 2 of life than in infants with negative C-reactive protein. Blood culture were only positive in infants with positive C-reactive protein can be regarded as an early marker for neonatal bacterial infection due to premature rupture of membranes and /or amnionitis.

Winkler M, Ruckhaberle KE, Baumann L, Schroder R, Schiller E. (1987) investigated the maternal serum levels of C-reactive protein (CRP) in 88 pregnancies with comparable gestational ages at the onset of premature birth symptoms. We found certain associations between possible prolongation of gestation by tocolysis and absence or presence of pathologic values of this acute-phase-protein. Positive CRP-values are associated with significantly lowered prolongation of pregnancy by tocolysis and subsequently lowered gestational age at birth.

Chaaban M, Jauniaux E, Nasreddine S, Jabry S, Duchateau J, Wilkin P. (1988) retrospectively studied the changes in the level of C-reactive protein (CRP) and of white blood cells in 82 patients who had premature rupture of the membranes between the 20th and the 36th week of pregnancy in order to estimate the possibility of prenatal screening for amnion infections in early rupture of the membranes. The level of CRP was shown to be quickly and significantly raised in cases of clinical or histological chorioamnionitis, whereas the change in maternal leucocytes alters little and later. The level of CRP can be worked out as an early biological marker which is sensitive and cheap in the clinical supervision of cases with early rupture of the membranes.

Kornman L, Jacobs V, Hodgson RP, Godfrey J, Dunlevy L, Tyler JP, Baird PJ, Hudson CN. (1988) designed a study to derive the predictive value of C-reactive protein (CRP) in peripheral venous serum of patients admitted to hospital with suspected premature rupture of the membranes (PROM). CRP was assayed by each of 4 separate methods and the results have been compared for accuracy and practical value with respect to clinical outcome and the histopathology of the placenta. Of the 4 techniques used only the latex test had characteristics suitable for a diagnostic screen. While the results were only semiquantitative, when comparisons were made to other techniques no significant change in clinical diagnosis would have been made. The results have confirmed that chorioamnionitis and preterm labour are often associated, but in some instances the extent of inflammatory infiltration was greater than might have been expected from the short time interval between documented membrane rupture and delivery. Thus it may be speculated that some cases of

PROM are secondary to, rather than causative of, infection. Finally it is suggested that a controlled therapeutic trial of active intervention in those cases of PROM with elevated CRP in the absence of other clinical parameters suggestive of intrauterine infection should be undertaken.

Hirsch W, Koppitz D, Morack G, Gerhardt C. (1989) For diagnosis of ascending intra-uterine infections, regular controls of the serum CRP levels were carried out in 129 patients with premature rupture of membranes and impending premature labour. The height of the maternal CRP level and the simultaneously determined leucocyte counts and band counts were compared with the peripartal fever morbidity (intrapartal fever, puerperal fever). It was established that patients who had a prepartal CRP level of 10 mg/l or more suffered febrile complications more frequently (fever morbidity 18.2%) than females in whom such high CRP values did not occur (fever morbidity 3.4%).

Moretti M, Sebai MB et al (1988) found that labor started within 24 hours of premature rupture of membranes in 81% of patients carrying babies larger than 2.5 kg. The situation is markedly different when premature rupture of membranes occurs early in gestation. In this latter case only 48% patients develop labor within 3 days after premature rupture of membranes.

Ibarra Chavarria V, Sanhueza Smith P, Mota Gonzalez M, del Rey Pineda G, Karchmer S. & associates (1989) studied correlation of the C-Reactive Protein levels (CRP) determined by nephelometric technique, with other infection indicators, and its

exactness in early detection of chorioamnioitis. Thirty patients were prospectively studied with pregnancies from 28 to 35 weeks of gestation with diagnosis of premature rupture of membranes (PROM); and were compared to control group (30 patients) with similar gestation without PROM, infection, autoimmune diseases or chronic inflammation. The value for CRP was 2 mg/dl. The study group included 17 patients considered as positive, and 13 negative; the differences in CRP values in infected women was significant and not infected ones with a probability less than 0.001 (Fisher), with a sensitivity of 94.12%, and specificity of 100% positive predictive value of 100%, and a negative predictive value of 98.86%. The present data show that CRP is an early detector of amniotic infection.

Teichmann AT, Arendt P, Osmers R, Speer CP. (1989) determined serum concentrations of leucocytes, C-reactive protein (CRP) and elastase-alpha 1-proteinase inhibitor (E alpha 1 PI) in 85 women during pregnancy and after birth to assess their diagnostic value in case of amniotic infections (AI). In ten patients clinically diagnosed AI could be confirmed by histopathological examination, three patients who fulfilled the clinical criteria showed no histological signs of infection. E alpha 1 PI-levels were found to be elevated to over 200 meg/l in six patients with clinical and histological infection and remained below this value in all other cases whereas leucocyte- and CRP concentrations raised in five out of these eight women but also showed false positive values in patients without AI. The application of betamethasone led to a marked elevation of leucocyte concentration. CRP levels were raised substantially after birth, whereas E alpha 1 PI remained unchanged. It was suggested that all three parameters should be taken into

consideration to increase diagnostic reliability in case of suspected amniotic infections.

CRP rises quickly after an inflammatory event and returns to normal within a week while the ESR rises slowly in response to increasing production of fibrinogen by the liver and falls slowly as well (Kushner I., 1990).

Kurki T, Teramo K, Ylikorkala O, Paavonen J. & associates (1990) studied the usefulness of maternal C-reactive protein (CRP) measurements in the diagnosis of chorioamnionitis and puerperal and neonatal infectious morbidity among 147 patients with preterm rupture of the membranes (PROM). Thirty-three patients developed chorioamnionitis, 10 patients developed puerperal endometritis, and 21 newborn infants developed neonatal infections. There was no difference in the highest antepartum CRP between patients with or without chorioamnionitis. The overall test performance for CRP was poor suggesting that elevated antepartum CRP may be misleading in the diagnosis of chorioamnionitis. However, use of serial CRP measurements increases the test performance. The high negative predictive value suggests that CRP is useful in predicting the absence of chorioamnionitis.

De Villiers WJ, Louw JP, Sirachan AF, Etsebeth SM, Shephard AF, De Beer FC (1990) measured serum level of C-reactive protein prospectively in normal pregnant women, newborn infants and women with preterm premature rupture of the membranes, focusing on the peripartum period. C-reactive protein level in 50 healthy women at 38 weeks gestation did not differ significantly

from previously established normal values. C-reactive protein levels in 67 healthy women sampled serially in labour from admission to 96 hours postpartum confirm the physiological occurrence of a major acute phase response. The serial C-reactive protein of 16 women with premature rupture of membranes did not differ significantly from the wide range of C-reactive protein levels found in normal postpartum period. This complicates the use of C-reactive protein as an early predictor of clinical chorioamnionitis.

M. Suri, S. Thirupurum, V.K.Sharma (1990) measured serum level of C-reactive protein serially in 25 healthy and 20 septicemic neonates and then compared with early diagnostic aids and prognostic indicators in this illness. Compared to healthy controls, septicemic neonates had significantly higher mean serum level of C-reactive protein ($p<0.01$). Neonates with septicemia, who recovered, had higher mean C-reactive protein levels than the group that died ($p<0.05$). As an early diagnosis aid C-reactive protein had a low you den index, whereas for prognosis its index was higher.

Ohlsson A, Wang E. (1990) critically reviewed published studies regarding sensitivity, specificity, and positive and negative predictive values of antenatal tests to diagnose chorioamnionitis or fetal-neonatal sepsis in preterm premature rupture of the membranes. Single, small studies, the precision of which has never been tested, showed good indices for repeatedly increased serum levels of C-reactive protein (greater than 20 mg/L), a high level of C-reactive protein greater than 40 mg/L, or a day-to-day coefficient of variation for C-reactive protein of greater than 30% in the prediction of histologic or clinical chorioamnionitis.

Teichmann AT, Arendt P, Speer CP. (1990) determined White blood cell count (WBC), C-reactive protein (CRP) and elastase alpha 1-proteinase inhibitor complex (E alpha 1 PI) in 85 women during pregnancy and after birth to assess their diagnostic value in case of amniotic infection syndrome (AIS). In ten patients clinically diagnosed AIS could be confirmed by histopathological examination, five patients who fulfilled the clinical criteria showed no histological signs of infection. E alpha 1 PI levels were found to be elevated to above 200 micrograms/l in nine patients with clinical and histological infection and remained below this value in all but one of the cases not showing signs of AIS. On the other hand, CRP concentrations were elevated in five out of these ten women, but also showed false-positive values in patients without AIS; leucocyte counts above 15,000/mm^3 have been observed in only one case before delivery. The application of betamethasone led to a marked elevation of leucocyte concentrations. CRP levels were raised substantially after birth, whereas E alpha 1 PI remained unchanged under both conditions. It was suggested that all three parameters should be taken into consideration to increase diagnostic reliability in case of suspected amniotic infections.

Passloer HJ. (1990) examined 30 cases of cervical incompetence in pregnancy morphological and infectious factors of the amniotic membranes with regard to a predictive function versus premature rupture of membranes (PROM). The sonographically visible chorioamniotic dissociation (CAD, cut-off-value greater than or equal to 3 mm) and the C-reactive protein (CRP, cut-off-value greater than or equal to 6 mg/l) showed a sufficient predictive force. The combination of CAD and CRP could predict all cases of PROM.

In addition, it could be shown that ascending infections are not only cause of PROM but also of morphological changes of the cervix and the amnial membranes in cervical incompetence. So therapy of an incompetent cervix should include competent vaginal sanitation.

Krohn M, Pahnke VG, Albrecht K, Trams G. (1990) evaluated the predictive diagnostic value of CRP in a wait-and-see management of PROM 110 courses of labour and childbed with 114 babies were investigated with respect to both complications of mother and newborn. Complications during childbed could only be seen in 2.7%, 3.4% of the newborns suffered from septic disease. With regard to complications of newborns and mothers mediated by infection, CRP was demonstrated to have a prepartum sensitivity of 79% in combination with a specificity of 80%. The absolute highest sensitivity and specificity of CRP were found at a cut-off point of about 14 mg/l, which is clearly above the normal level of 6 mg/l serum. In almost 50% of the cases with maternal antepartum CRP levels up to 20 mg/l no pathogenic bacterial invasion could be revealed in the newborn. CRP therefore can be taken as an additional parameter in the management of PROM even in cases treated with corticosteroids due to its specificity and indicates better than WBC and polymorphs when to terminate the wait-and-see attitude.

He JP. & Associates (1990) studied Sensitivities of C-reactive protein (CRP) and acute-phase proteins (APP) in predicting the infection induced by PROM. The results showed that CRP could demonstrate clinical or subclinical infection with a sensitivity of 100% and 86.7% respectively, and no false positivity was observed. CRP increased remarkably at least 24 hours earlier than that of other

parameters when infection occurred. On the other hand the sensitivity of APP (64.1%) was lower with false positivity. Thus, CRP may be used in clinical practice to predict infection resulted from PROM during conservative management.

Pourcyrous M, Bada HS, Korones SB, Barrett FF, Jennings W, Lockey T. (1991) evaluated the C-reactive protein (CRP) level in 142 infants requiring investigation for suspected infection. After excluding two neonates because of incomplete data, there remained 140 neonates, of whom 16 had septicemia. Fifteen of 16 had increased CRP levels. The CRP value was not elevated in any baby (n=5) who had positive blood cultures for Staphylococcus epidermidis, all of whom had an uneventful clinical course. The CRP level was elevated in all six babies with meconium-aspiration syndrome, but was normal in five infants whose viral cultures were positive. Ninety-nine percent of uninfected babies had normal CRP values. Overall, CRP was a valuable test for diagnostic confirmation of bacterial infection. Elevated CRP level was always accompanied by at least one abnormality in the other tests performed. Although the study was not intended to predict clinical onset of bacterial disease, our results suggest that the CRP level, because of a high negative predictive value, may be useful in ruling out bacterial infection.

Young B, Gleeson M, Cripps AW. (1991) had reviewed the literature to determine the value of C-reactive protein (CRP) measurements in the diagnosis and management of a wide range of conditions. CRP levels are of value in 6 clinical situations: (a) monitoring the response to antibiotic treatment in patients with known bacterial infections, (b) in obstetric patients with premature

rupture of membranes, a rise in CRP can give early warning of intrauterine infections, (c) differentiation between active disease and infections in patients with systemic lupus and ulcerative colitis where the level of response to active disease has been previously established, (d) as a measure of disease activity and response to disease-modifying drugs in rheumatoid arthritis, (e) early detection of complications in postoperative patients, (f) in differentiating between infection and graft-versus-host-disease in bone marrow transplant patients. CRP levels have been used in an attempt to differentiate between bacterial and viral infections in various clinical situations, however the published literature does not support this role.

Berardi JC, Hutin S, Godard J, Madinier V, Delanete A, Berardi-Grassias L. (1991) & associates compared the sensitivity and ability to predict the onset of chorio-amnionitis of the conventional clinical signs (hyperthermia, fetal tachycardia, discolored amniotic fluid) and paraclinical signs (hyperleukocytosis and bacteriology of the amniotic fluid) with those of the assay of C reacting protein in the maternal plasma in context of premature rupture of the membranes. The latter test is apparently more sensitive and of greater positive predictive value in this disorder.

Mueller-Heubach E, Rubinstein DN, Schwarz SS. Examined the placentas of 1843 deliveries for the presence of histologic chorioamnionitis, which was classified as mild, moderate, or severe. Chorioamnionitis was present in 7.5% of patients who underwent cesarean before labor and in 18 and 32% of those delivering at term and preterm, respectively. Chorioamnionitis was severe in 74% of

preterm but in only 15% of term deliveries. Premature rupture of membranes (PROM) was more frequent with preterm than with term delivery, with chorioamnionitis present in 42 and 15% of patients, respectively. Although chorioamnionitis was equally frequent in women with intact membranes delivering preterm and term, chorioamnionitis was severe in 63% of preterm and 14% of term deliveries (P less than .001). The frequency and severity of chorioamnionitis were related inversely to gestational age at preterm birth. Preterm delivery was more frequent in black than in white patients (19 versus 9%) and in indigent clinic versus private patients (13 versus 7.5%). However, there was no significant difference in frequency and severity of chorioamnionitis between black and white or between indigent clinic and private patients who delivered preterm. Among term births, chorioamnionitis was more often severe in black than in white patients. Chorioamnionitis in term deliveries was more frequent in clinic than in private patients; however, this was not true when only severe chorioamnionitis was considered. There were no differences in PROM between these patient populations. Thus, higher preterm birth rates in black and indigent clinic populations are not due to the more frequent occurrence of chorioamnio-nitis.

Watts DH, Krohn MA, Hillier SL, Eschenbach DA. (1992) evaluated the relationships between gestational age, neonatal outcome, and amniotic fluid (AF) bacteria, they obtained AF from women with intact membranes in idiopathic preterm labor. Positive cultures were obtained from 20 (19%) of 105 women. The frequency of positive cultures was inversely related to gestational age: 23-26 weeks, nine of 20; 27-30 weeks, four of 24; and 31-34 weeks, seven

of 61 (chi2 for trend, P less than .001). Fusobacterium nucleatum, Bacteroides ureolyticus, and Ureaplasma urealyticum were the most common isolates. Facultative and anaerobic bacteria were more commonly isolated from women at less than 30 weeks' gestation, and Ureaplasma urealyticum was commonly isolated at greater than 30 weeks' gestation. Forty percent of the patients identified as having positive AF facultative and anaerobic cultures by the research laboratory had negative cultures in the clinical laboratory. Clinical characteristics and maternal white blood cell count and differential did not differ between women with and without positive cultures. Elevated C-reactive protein levels and a positive AF Gram stain were the two most sensitive and specific methods to predict positive AF cultures. Women with positive cultures delivered a median of 1.0 day after enrollment, compared with 28.5 days for women with negative cultures. The median gestational age at delivery for women with positive cultures was 27.5 weeks, and the median birth weight was 866 g. Positive AF cultures were associated with respiratory distress syndrome, broncho pulmonary dysplasia, and neonatal death. If occult AF infection among women in preterm labor is a treatable cause of preterm birth, then treatment could markedly reduce both perinatal morbidity and mortality.

Russell GA, Smyth A, Cooke RW. (1992) Compared serial neutrophil band cell counts with C reactive protein measured by rate nephelometry. The 'gold standard' was a positive culture and the performance of the tests was compared by the technique of receiver operating characteristics (ROC) as well as sensitivity and specificity. A total of 172 septic screens were performed in 56 patients. The operational diagnostic cut off values were: C reactive protein greater

than 8 mg/l, immature:total neutrophil ratio (I:T ratio) greater than 0.2, and band count greater than 5%. Compared with the sensitivity of C reactive protein (71%), I:T ratio (34%) was significantly different but band count (69%) was not. The specificity of C reactive protein (72%) was better than band count (39%) .ROC curves were constructed for all possible diagnostic cut off values of the tests and superior performance was demonstrated for C reactive protein compared with band count and I:T ratio. They concluded that C reactive protein is a useful early indicator of infection in neonates and that ROC curves permit comprehensive and graphic comparison between tests and the calculation of optimal diagnostic cut off values.

Pourcyrous M, Bada HS, Korones SB, Baselski V, Wong SP. (1993) performed study to determine prospectively whether, in the presence of proved or presumed bacterial infection, the sensitivity of serum C-reactive protein (CRP) response could be enhanced by serial rather than single determinations. Thay also sought to assess CRP responses to clinically identified noninfectious disorders. The CRP responses of 491 infants on 691 occasions of suspected infection were assessed. CRP levels were measured initially and twice again at 12-hour intervals (rate immunonephelometry). Assessments also included a blood culture, complete blood cell count, and chest radiograph and culture of spinal fluid when appropriate. CRP responses were correlated with four designated clinical groups: (1) positive blood or cerebrospinal fluid cultures (n = 190); (2) negative blood culture-definite infection (necrotizing enterocolitis stages 2 and 3, pneumonia, subcutaneous abscess) (n = 52); (3) negative blood culture-possible infection (antenatal risk factors, meconium

aspiration, positive urine group B streptococcus antigen, necrotizing enterocolitis stage 1, febrile infants) (n = 287); and (4) negative blood culture-no infection (respiratory distress syndrome, transient tachypnea of the newborn, patent ductus arteriosus, tissue trauma) (n = 160). Diagnoses were made before CRP results were known. In all, 187 (27%) of the blood cultures were positive. A single organism was recovered from 174 of these; two organisms from 13. Among the single-organism cultures, 50 (29%) were Gram-negative, 120 (69%) were Gram-positive, and 4 (2%) were budding yeasts. CRP levels were elevated in various groups as follows: in the positive blood culture group (by organism), Gram-negative rods, 92% (46/50); group B streptococcus, 92% (12/13); Staphylococcus aureus, 89% (8/9); group D streptococcus, 71% (10/14); Streptococcus viridans, 60% (6/10); Staphylococcus epidermidis, 55% (40/73). In the negative blood culture-definite infection group, CRP levels were abnormal in 88%; in the negative culture-possible infection group, CRP was elevated in 33%; and in the negative blood culture-no infection group, CRP was elevated in 9%. Serial determinations of CRP resulted in enhanced sensitivity in the positive blood culture group, the negative blood culture-definite infection group, and the negative blood culture-possible infection group. Initial determinations by themselves were inadequately sensitive. Serial determinations did not enhance sensitivity of the negative blood culture-no infection group. High specificity (91%) is suggested by the low incidence of abnormal CRP levels among infants who were not infected. These data suggest that it would be appropriate to conduct a cautious, controlled trial to assess the safety of discontinuing antibiotic therapy if three serial CRP measurements are normal and if there are no other clinical factors suggestive of

infection. The data also indicate the necessity for serial determinations of CRP for optimal sensitivity.

Beck T, Bahlmann F, Weikel W. (1993) Investigated the relationship between histologically confirmed chorioamnionitis and maternal and fetal inflammation parameters in 69 patients on the basis of inflammation of the membranes, placenta and cord occurring in histomorphologic stages. Their results show the C-reactive protein to be a sensitive and specific indicator of chorioamnionitis and closely correlated with both the histologic stage and the severity of the chorioamnionitis. They therefore advocated adoption of the histologic result as the "gold standard" for evaluating subclinical and clinically manifest forms of intrauterine infection.

Mazor M, Zitzer P, Chaim W, Maymon E, Kuperman O & associates (1993) found that there is a strong association between systemic and intrauterine infection and preterm delivery. C-reactive protein (CRP) is considered a nonspecific marker for intrauterine inflammation. They determined its blood level in 100 women who presented with preterm labor and intact membranes and delivered prematurely. CRP levels were used as a noninvasive marker for infection to compare clinical characteristics between women who delivered prematurely at 31-36 and 24-30 weeks of gestation. Women who delivered earlier than the 24th week of gestation had a higher rate of positive CRP levels than those who delivered later, 54% vs. 24%, respectively ($p < 0.0001$). Moreover, women from the lower gestational age group with positive CRP levels had significantly different clinical characteristics than those in the same group but with negative CRP levels. There was a significant difference between

Jewish and Bedouin women in cervical dilatation and time interval from hospitalization to delivery between those with positive CRP and those with negative CRP levels. They conclude that patients who delivered prematurely at 24-30 weeks had higher rates of an inflammatory etiology than women who also delivered prematurely, but at a more advanced gestational age.

Mazor M, Kassis A, Horowitz S, Wiznitzer A, Kuperman O, Meril C, Glezerman M. & associates (1993) determined the relationship between C-reactive protein (CRP) levels and intraamniotic infection in 48 women presenting with preterm labor and intact membranes. Blood samples for CRP tests were obtained immediately before the performance of transabdominal amniocentesis. The prevalence of amniotic fluid cultures positive for organisms was 14.6%. In 16 women (33.3%) positive CRP levels were obtained. There were no significant differences in the prematurity rate or the prevalence of microbial invasion of the amniotic cavity between women with positive CRP levels and women with negative levels. The sensitivity, specificity, and positive and negative predictive values for the detection of amniotic infection were 71.5%, 73.2%, 31.3% and 93.8%, respectively. Based on these results, they suggested that in women with preterm labor and negative CRP levels, routine amniocentesis might not be essential to the initial workup.

Watts DH, Krohn MA, Hillier SL, Wener MH, Kiviat NB, Eschenbach DA. & Associates (1993) evaluated clinical, microbiologic, and histologic findings associated with elevated C-reactive protein levels among women in preterm labor or with

preterm premature rupture of the membranes (PROM). Obstetric data, serum C-reactive protein levels, and amniotic fluid (AF) and chorioamniotic membrane cultures and histology were obtained on 203 women presenting between 22-34 weeks' gestation in preterm labor or with PROM. Women with C-reactive protein greater than 1.5 mg/dL were more likely to deliver within 7 days of enrollment (54 of 68, 79%) than were women with normal C-reactive protein levels (45 of 135, 33%) ($P < .001$). The median C-reactive protein levels and association with rapid delivery did not differ between women with intact versus ruptured membranes. Elevated C-reactive protein levels were associated with a positive AF culture among women in preterm labor with intact membranes. To control for confounding by a long interval to delivery, only the group delivering within 7 days was considered for evaluation of C-reactive protein levels and placental and infant outcome. Among women delivering within 7 days, elevated C-reactive protein was associated with the development of clinical chorioamnionitis and with infant death before hospital discharge, but not with a positive membrane culture or histologic chorioamnionitis. Elevated C-reactive protein appears to be associated with AF infection, delivery within 7 days of admission, and infant death among women delivering preterm, but not with membrane infection or inflammation. Elevated C-reactive protein may be helpful in determining the need for AF culture and in targeting studies of antibiotic therapy among women in preterm labor or with preterm PROM.

Thompson PJ, Greenough A, Davies E, Nicolaides KH. & Associates measured C-reactive protein (CRP) in fetal blood obtained by cordocentesis from 17 patients with preterm prelabour rupture of

the membranes (PPROM). CRP was detected in the blood of eight of the 17 fetuses. Six fetuses, five of whom may have been infected had CRP > or = 0.8 mg dl-1. The remaining 11 fetuses as well as 25 healthy term infants who had cord blood taken immediately post delivery had CRP < 0.6 mg dl-1. These results suggest that elevation of fetal CRP levels may be a useful indicator of fetal infection in pregnancies complicated by PPROM.

Da Silva, et al 1995 reviewing the use of CRP as a tool for diagnostic neonatal sepsis, concluded that CRP is probably the best available diagnostic test. Further, Yentis SM, Soni N, Sheldon JC, 1995 found daily measurements of CRP to correlate with resolution of sepsis, specifically, A decrease in CRP by 25% or more from previous days level was a good indicator of resolution of sepsis, with a sensitivity of 97%, specificity of 95% and predictive value of 97%.

Schouten-Van Meeteren NY, Rietveld A, Moolenaar AJ, Van Bel F. (1995) C-reactive protein rises in blood in an acute-phase response in adults, children, and neonates. In a prospective study of the influence of perinatal asphyxia, premature rupture of membranes, hyperbilirubinemia, and respiratory distress syndrome on levels of C-reactive protein in the neonate, we detected no confounding effect on the rise of C-reactive protein level in infants with these pathologic perinatal conditions, as compared with the results of a control group.

Luttkus A, Windel K, Dudenhausen JW. & Associates (1995) in an open prospective investigation determined the median levels of C-reactive protein in abnormal collective. The median lies at 0.8 mg/dl in maternal serum withdrawn sub partu and in umbilical vein

blood, and at 1.45 mg/dl immediately after ligation of the cord. Hopes on finding a biochemical parameter that could supply reliable information already during labour on a possible inflammatory infection in mother and child, did not materialize from the data found. Prediction of an infection of the newborn using CPR is only minimal. The most important practical information is given by the negative CRP in the umbilical vein blood. In this case the probability of an infection of the newborn is very slight.

Yoon BH, Jun JK, Park KH, Syn HC, Gomez R, Romero R. & associates (1995) compared the diagnostic performance of maternal blood C-reactive protein, white blood cell count (WBC), and amniotic fluid (AF) WBC in the identification of positive AF culture, histologic and clinical chorioamnionitis, and neonatal morbidity in women with preterm premature rupture of membranes (PROM). Maternal blood was collected for the determination of C-reactive protein and WBC at the time of amniocentesis from 90 women with preterm PROM. Amniotic fluid was cultured for aerobic and anaerobic bacteria as well as mycoplasmas. Amniotic fluid WBC was determined for research purposes. Receiver operating characteristic curve and logistic regression were used for statistical analysis. The prevalence of positive AF culture was 28% (25 of 90). Women with positive AF culture and clinical chorioamnionitis had significantly higher median C-reactive protein, WBC, and AF WBC than did women ,without these conditions ($P < .05$), whereas women with histologic chorioamnionitis and significant neonatal morbidity had higher median C-reactive protein and AF WBC, but not WBC, than those without the conditions ($P < .05$). An AF WBC of at least 20 cells per mm3 had a greater sensitivity than C-reactive protein

cutoff, 0.7 mg/dL) and WBC (cutoff, 13,000 cells per mm3) in the detection of positive AF culture and histologic chorioamnionitis. Logistic regression analysis indicated that among AF WBC, C-reactive protein, and WBC, AF WBC was the best predictor of positive AF culture (odds ratio [OR] 24.2, 95% confidence interval [CI] 6.0, 97.5, $P < .001$), histologic (OR 74.0, 95% CI 7.4, 736.3, $P < .001$) and clinical chorioamnionitis (OR 8.9, 95% CI 0.9, 85.6, $P = .057$), and neonatal morbidity (OR 4.3, 95% CI 1.1, 16.6, $P < .05$).

Berger C, Uehlinger J, Ghelfi D, Blau N, Fanconi S. (1995) prospectively compared the diagnostic value of C-reactive protein (CRP) and white blood cell counts for detection of neonatal septicaemia. Sensitivity and specifity in receiver operating characteristics, and positive and negative predictive value of CRP and white blood cell count were compared in 195 critically ill preterm and term newborns clinically suspected of infection. Blood cultures were positive in 33 cases. During the first 3 days after birth CRP elevation (sensitivity 75%, specifity 86%), leukopenia (67%/90%), neutropenia (78%/80%) and immature to total neutrophil count (I/T) ratio (78%/73%) were good diagnostic parameters, as opposed to band forms with absolute count (84%/66%) or percentage (79%/71%), thrombocytopenia (65%/57%) and toxic granulations (44%/94%). Beyond 3 days of age elevated CRP (88%/87%) was the best parameter. Increased total (84%/66%) or percentage band count (79%/71%) were also useful. Leukocytosis (74%/56%), increased neutrophils (67%/65%), I/T ratio (79%/47%), thrombocytopenia (65%/57%) and toxic granulations had a low specifity. The positive predictive value of CRP was 32% before and 37% after 3 days of age, that of leukopenia was 37% in the first 3 days. During the first 3

days of life CRP, leukopenia and neutropenia were comparably good tests while after 3 days of life CRP was the best single test in early detection of neonatal septicaemia. Serial CRP estimations confirm the diagnosis; monitor the course of infection and the efficacy of antibiotic treatment.

Kawamura M, Nishida H. (1995) evaluated serial changes in C-reactive protein values in 108 term and 240 preterm newborn infants with suspicion of infection, and the changing patterns of C-reactive protein values were compared with clinical outcome. For a diagnosis of infection, the negative predictive values in term and preterm infants were 99.0% and 97.8%, respectively, although the sensitivities were 61.5% and 75.0%, respectively. Antibiotic therapy was started at birth and discontinued when the changing pattern of C-reactive protein and clinical findings did not suggest infection. As a result, mean durations of administration of antibiotics in the term and preterm infants were 3 and 4 days, respectively. Recognition of the changing pattern of C-reactive protein was very useful in excluding infection and minimizing unnecessary antibiotic therapy in managing neonatal infection.

Yoon BH, Yang SH, Jun JK, Park KH, Kim CJ, Romero R. & associates (1996) compared the diagnostic and prognostic performance of maternal blood C-reactive protein, white blood cell count (WBC), and temperature with that of amniotic fluid (AF) WBC in preterm labor. One hundred two women with preterm labor and intact membranes were studied. Maternal blood was collected to measure C-reactive protein concentration and WBC, and maternal temperature was also measured. Amniotic fluid obtained by

amniocentesis was cultured and WBC determined. Receiver operating characteristic curve, logistic regression, and survival techniques were used for analysis. Patients with acute histologic chorioamnionitis had significantly higher median C-reactive protein concentration, WBC, temperature, and AF WBC than patients without this lesion ($P < .05$). Receiver operating characteristic curve and survival analysis demonstrated that an elevated C-reactive protein, WBC, or AF WBC was strongly associated with the likelihood of histologic chorioamnionitis, shorter interval to delivery, clinical chorioamnionitis, and neonatal morbidity ($P < .05$ for each). Of all the tests, AF WBC was the best independent predictor of a positive AF culture (odds ratio [OR] 16.8), interval to delivery (hazard ratio 5.7), clinical chorioamnionitis (OR 15.2), neonatal sepsis (OR 16.8), and significant neonatal complications (OR 7.4), after other confounding variables were adjusted ($P < .05$ for each). An elevated C-reactive protein, WBC, or AF WBC identified patients with intrauterine infection and adverse perinatal outcomes.

Nowak M, Oszukowski P, Szpakowski M, Wladzinski J, Kaminski T, Malinowski A. (1998) evaluated the influence of dexamethasone administration on selected markers of infection during the expectant management of premature rupture of membranes (PROM). A group of 80 patients with PROM before 35 weeks' gestation were evaluated prospectively and managed expectantly. They applied the expectant management with the permanent use of tocolysis, antibiotics, steroids (4 mg of dexamethasone every 8 hr for 2 days, every week), amnioinfusions of artificial amniotic fluid and intravaginal chemotherapeutics. Patients were monitored with frequent vital signs, fetal heart rate evaluation and everyday blood

tests as follows: C-reactive protein (CRP), white blood cell count (WBC) and erythrocyte sedimentation rate (ESR). After completing 25 weeks antenatal fetal surveillance included a nonstress test at least once a day. All afterbirths were examined to establish the presence of histologic chorioamnionitis. WBC raised significantly in the first day of steroidotherapy (mean: 14,895/mm3), was elevated in the second day (15,716/mm3) and the day after (15,100/mm3), and decreased to the normal limit in the second day after steroids (12,316/mm3); $p < 0.001$. There were no statistically significant differences in the results of CRP and ESR connected with the administration of dexamethasone. Dexamethasone administration caused the independent of infection and temporary elevation of WBC. They did not observe such influence on the results of CRP and ESR.

Nowak M, Oszukowski P, Szpakowski M, Malinowski A, Maciolek-Blewniewska G. & associates (1998) analyzed the efficacy of serum C-reactive protein (CRP), white blood cell count (WBC) and erythrocyte sedimentation rate (ESR) serial evaluations in the prediction of chorioamnionitis in cases of premature rupture of membranes (PROM).

A group of 80 patients with PROM before 35 weeks' gestation were evaluated prospectively and managed expectantly. We applied the expectant management with the permanent use of tocolysis, antibiotics, steroids, amnioinfusions of artificial amniotic fluid and intravaginal chemotherapeutics. Patients were monitored with frequent vital signs, fetal heart rate evaluation and everyday blood tests as follows: CRP, WBC and ESR. All afterbirths were examined to establish the presence of histologic chorioamnionitis (gold

standard of intrauterine infection). 59 (73.7%) patients had significant chorioamnionitis on histopathology and only 15 of them had clinical chorioamnionitis. Serum CRP serial determinations (definition of abnormal tests: 1) > 1.2 mg/dl; 2) > 2.0 mg/dl; 3) > 1.2 mg/dl and increasing in two consecutive days) were found the most reliable with a sensitivity 1) 91.5%; 2) 85%; 3) 88%, specificity 57%; 76%; 86%, positive predictive value 86%; 90%; 94.5%, negative predictive value 70.5%; 64%; 72% and accuracy 82.5%; 82.5%; 87.5% respectively. The efficacy of WBC (abnormal tests: > 12500/mm3; > 15000/mm3; > 12500/mm3 and increasing in two consecutive days) and ESR (abnormal tests: > 60 mm/h; > 60 mm/h and increasing in two consecutive days) serial evaluations was significantly lower. Moreover, in cases of chorioamnionitis CRP increased above the upper limit of normal 3 days earlier than WBC or ESR. CRP was found the most reliable indicator of histologic chorioamnionitis and indicated the presence of intrauterine infection earlier than WBC or ESR.

CRP binds to apoptotic cells, protects the cells from assembly of the terminal complement components, and sustains an anti-inflammatory innate immune response (Gershov D et al 2000).

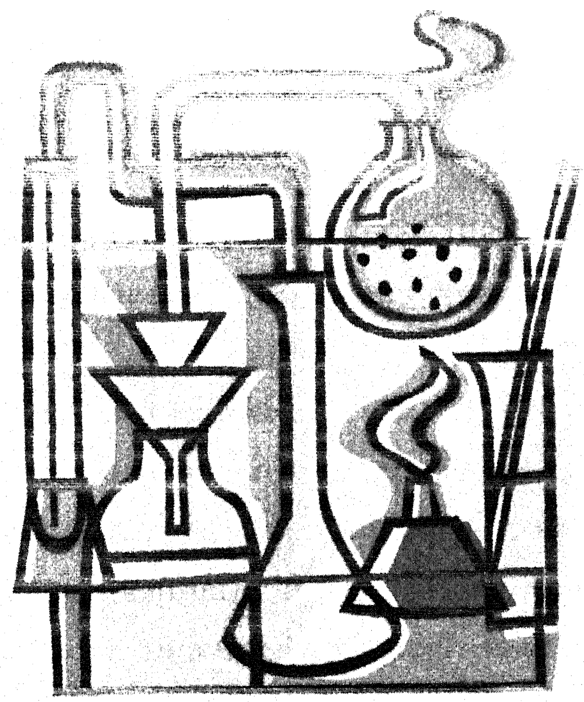
Ghezzi F, Franchi M, Raio L, Di Naro E, Bossi G, D'Eril GV, Bolis P. (2001) investigated whether the amniotic fluid C-reactive protein level at the time of genetic amniocentesis is a marker for spontaneous preterm delivery before 34 and 37 weeks of gestation. Women who underwent genetic amniocentesis between 15 and 18 weeks of gestation with (1) singleton gestation, (2) uneventful pregnancy course before the amniocentesis, and (3) absence of fetal

bnormalities were included in the study. Patients with abnormal aryotype were excluded. C-reactive protein concentration was neasured in amniotic fluid and in maternal blood immediately after genetic amniocentesis. All patients were followed until delivery for he occurrence of pregnancy complications. Nonparametric tests and receiver-operating characteristic curve analysis were used for statistical purposes. The prevalence of spontaneous preterm delivery before 34 and 37 weeks was 3.3% (10 of 306 pregnancies) and 8.5% (26 of 306 pregnancies), respectively. Women with preterm delivery at <37 weeks had a higher median (range) of amniotic fluid C-reactive protein concentration than those women who delivered at term (median, 113.3 ng/mL [range, 16-623 ng/mL] vs median, 57.8 ng/mL [range, 0-808.9 ng/mL]; P <.005). Women with preterm delivery at <34 weeks had a higher median (range) amniotic fluid C-reactive protein concentration than those women who delivered at term (median, 183.8 ng/mL [range, 46.5-447 ng/mL] vs median, 57.8 ng/mL [range, 0-808.9 ng/mL]; P <.005]. No correlation was found between amniotic fluid C-reactive protein and maternal blood C-reactive protein concentrations. No relationship was found between maternal blood C-reactive protein concentration and preterm delivery before either 34 or 37 weeks. Amniotic fluid C-reactive protein concentration of >110 ng/mL had a sensitivity of 80.8% and a specificity of 69.5% in the prediction of spontaneous preterm delivery at <34 weeks. This study supports the theory that a subclinical intrauterine/fetal inflammatory process early in gestation may be important for the occurrence of preterm delivery in the second half of gestation.

Chen SU, Ko TM, Hwa HL, Lu PJ, Ho HN, Yang YS. (2001) nvestigated the diagnostic value of maternal serum C-reactive)rotein (CRP) in the recognition of chorioamnionitis in patients ındergoing fetal reduction. Seventy-one gravidas with high-order nultifetal pregnancies, including 46 with triplets, 18 with quadruplets, and 7 with quintuplets, who underwent transabdominal fetal reduction to twins during the 10th-14th gestational week were recruited. The subjects were followed up clinically and ultrasonographically 1 week and 1 month after fetal reduction for signs of infection, premature uterine contraction, and premature rupture of the membranes CRP levels were measured prior to fetal reduction and at follow-up examinations, and were compared. Among the 71 mothers, 65 (92%) were normal after fetal reduction. The CRP levels were not significantly different prior to the procedure (0.27 +/- 0.26 mg/dL), and 1 week (0.23 +/- 0.24 mg/dL) and 1 month (0.24 +/- 0.20 mg/dL) later. There was no correlation between the number of fetuses reduced and the CRP levels. Six (8%) experienced leakage of amniotic fluid after fetal reduction. Three patients had normal CRP levels at that time and at the following tests. The pregnancies continued smoothly after conservative treatment. The other three patients had elevated CRP levels when leakage of amniotic fluid occurred. Fever and uterine irritability developed subsequently despite parenteral antibiotics and tocolytic therapy. Daily checks showed increasing CRP levels. The pregnancies were aborted, and the histology of the placental membranes revealed chorioamnionitis with infiltration of acute inflammatory cells. The absorption of inactive gestational tissue after fetal reduction did not affect CRP levels. CRP may be used as a marker of intrauterine infection after fetal reduction.

The acute-phase response, an important pathophysiologic phenomenon, replaces the normal homeostatic mechanisms with new set points that presumably contribute to defensive or adaptive capabilities. The functions of these changes are highly variable and diverse: some participate in initiating or sustaining the inflammatory process, others modulate it, and still others have adaptive roles. These changes are induced by a complex intercellular signaling system of which the chief constituents are inflammation-associated cytokines. Several cytokines, particularly interleukin-6, stimulate the production of acute-phase proteins in response to varied stimuli. The patterns of cytokine production and of the acute-phase response differ in different inflammatory conditions. Acute-phase changes reflect the presence and intensity of inflammation, and they have long been used as a clinical guide to diagnosis and management. For this purpose, determination of serum C-reactive protein has advantages over the traditional strategy of measuring the erythrocyte sedimentation rate.

MATERIAL & METHODS



MATERIAL AND METHODS

The study was conducted in the Department of Obstetrics and Gynaecology in cooperation with Department of Biochemistry in M.L.B. Hospital, M.L.B. Medical College, Jhansi since July 2002 to August 2003.

One hundred antenatal cases were selected from out patient Department and in-patient Department of Obstetrics and Gynaecology of M.L.B. Medical College Jhansi. The following samples were analysed.

- Forty cases of normal pregnancy between 28-40 weeks of gestation taken as controls.

- Sixty cases of confirmed premature rupture of membranes before 35 weeks of gestation taken in study group.

Detailed history was taken. A patient's description of leakage of amniotic fluid is one of most valuable clues for the diagnosis of PROM. History alone has an accuracy of 90%. A patient may describe a "gush of amniotic fluid," intermittent leaking of small amount of fluid, or increased perineal moisture. As pregnancy advances, urinary stress incontinence becomes more common and may be confused with symptoms of membrane rupture. Detailed history taking was followed by complete general and obstetrical examination of all cases.

Any associated medical illness including infection, Rheumatoid arthritis or Systemic Lupus Erythmatosus was excluded from the study.

PROM was confirmed by an alkaline pH on Nitrazine paper after a history suggestive of PROM was elicited. In uncertain cases a sterile speculum examination was performed in each patient to note the presence of liquor over the lower blade of Sims speculum before it came in contact of vaginal wall.

Liquor was confirmed by following: -

(a). Gross pooling of amniotic fluid in the posterior fornix.

(b). Positive ferning.

(c). Nitrazine test

(d). Evaporation test

FERN TEST: Ferning results from the drying out of salts contained (sodium chloride) in the amniotic fluid. To perform the test, a drop of amniotic fluid is placed on a glass slide and allowed to dry. In patients of less than 28 weeks of gestation, it is best to heat dry thick drops of fluid with matchstick. The preparation is observed under the microscope, looking for a crystallization pattern that resembles a fern.

The accuracy of the test is affected by blood or meconium because they prevent amniotic fluid arborization. Higher concentrations of blood alter the morphology of the crystals resulting in a "skeletonized" ferning pattern.

The test may produce false positive results if the sample is obtained from the cervix because dry cervical mucous forms an arborisation pattern that may be confused with premature rupture of membranes .In contrast to the delicate and discrete ferning of amniotic fluid, cervical mucus produces a thick, dark, wide arborization pattern.

Many studies showed the test to have a sensitivity of 96-99%, specificity of 96-98%, positive predictive value of 98-99%, and negative predictive value of 90-99%. Diagnosis of premature rupture of membranes is close to 100% reliable if vaginal fluid gives both positive nitrazine and positive fern test.

NITRAZINE TEST: The vaginal pH normally ranges between 4.5 to 5.5 whereas that of amniotic fluid is usually between 7.0 to 7.7. The use of indicator nitrazine for the diagnosis of ruptured membranes, first suggested by Baptisti (1938), is a simple and fairly reliable method. Nitrazine is an indicator paper impregnated with sodium dinitrophenylazonaphthol disulfonate.

The colour of the reaction is interpreted by comparison with a standard colour chart. The pH of vaginal secretion is estimated by inserting strile cotton tipped applicator deeply into vagina & then touching it to the strip of nitrazine paper & comparing the colour of the paper with the chart supplied with paper.

Table showing Color changes of Nitrazine paper

Colour	pH
Yellow	4.5
Olive yellow	5.0
Deep olive yellow	5.5
Olive green	6.0
Blue green	6.5
Blue gray	7.0
Deep blue	7.5

The membranes probably are intact if the colour of paper remains yellow or changes to olive yellow (pH 5.00-5.5) A pH >6.5 is consistent with ruptured membranes. In 1940, **Abe** found the sensitivity, specificity, and positive and negative predictive values of Nitrazine paper for the documentation of rupture of membranes to be 97%, 99%, 99% and 96%, respectively. False positive reactions may occur in 4-15% of cases due to alkalization of the vagina by blood, semen, soap, antiseptic solution, and infection with Trichomonas or bacterial vaginosis. Vaginal contamination with infected urine also may cause a false-positive result because urine (normally acidic) will undergo alkalization in the presence of Proteus and other urease-producing organism. False-negative reactions may occur in 0-7% of cases due to decreased efflux of amniotic fluid as the time between membrane rupture and testing increases.

EVAPORATION TEST:

Liquor is heated on a slide until the water content has evaporated if a white residue is left, amniotic fluid is present .If the residue is brown the membranes are intact.

Cervical swab was taken for culture from each patient of study group.

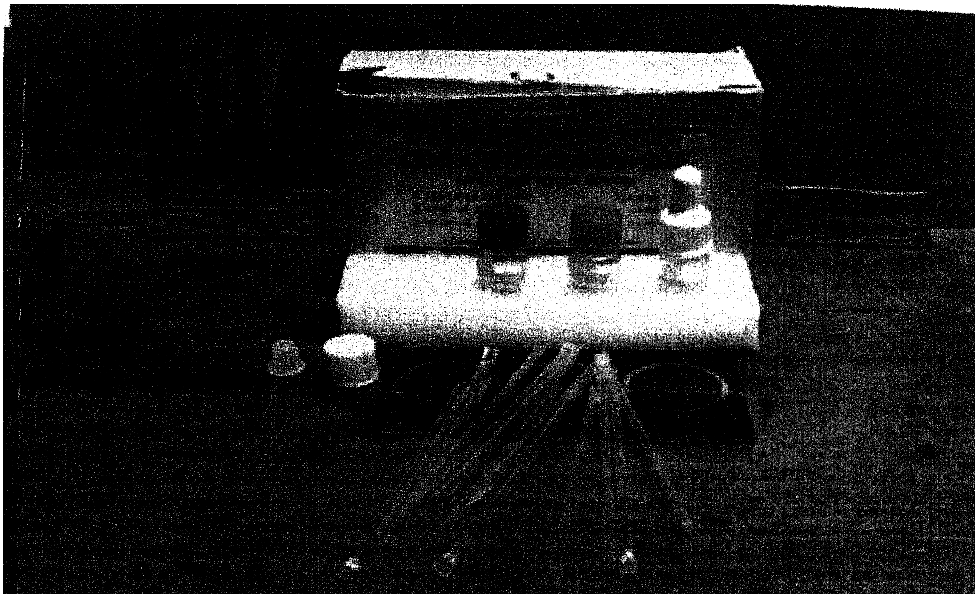
No digital pelvic examination was performed until active, irreversible labor ensued. Menstrual history was used to establish gestational age, and, if dates were uncertain, biparietal diameter estimations obtained by ultrasonography were used as the best estimate of gestational age.

MONITORING OF PATIENTS:

Daily monitoring before delivery: Parameters taken were - maternal pulse rate, maternal temperature, foetal heart rate, total leucocyte count, differential leucocyte count, ESR and daily C-reactive protein determination. Total leucocyte count was done by standard method using neubauer's chamber and was considered positive if total leucocyte count was in between 12000/cumm-15000/cumm. Differential leucocyte count was performed on a peripheral blood smear stained with Leishman's stain and polymorph, lymphocyte, eosinophils and basophils were identified.

ESRs were performed by the traditional Westergren method.

Fetal heart rate, maternal temperature, and uterine tenderness or contractions were evaluated every 8 hours. Most patients were managed expectantly. Corticosteroids were routinely administered (12 mg of betnesol, 2 doses, 12 hours apart). During the first 48 hours, whenever regular uterine activity developed, tocolytic therapy (isoxsuprine drip) was instituted. Conservative management was interrupted if labor occurred or if clinical evidence of



C-REACTIVE PROTEIN KIT

chorioamnionitis developed. Labor was induced if maternal temperature rose to 38°C or higher, if the uterus became tender and irritable, or if foul smelling amniotic fluid was noted, and if abnormal fetal heart rate developed.

Criteria used to diagnose clinical chorioamnionitis before delivery: -

1. Maternal fever more than 100.4 F
2. Tender and irritable uterus.
3. Foul smelling discharge per vaginum.
4. Abnormal foetal heart rate i.e. foetal tachycardia (>160 beats/minute).

DAILY C-REACTIVE PROTEIN DETERMINATION: Serial C-reactive protein determination was done in each patient. Blood sample was collected by venipunture. No special preparation of the patient was required prior to specimen collection. For the test only unhaemolysed, nonlipemic, non turbid serum was used.

C-reactive protein determination was done by using latex agglutination method with the help of C-reactive protein reagent kit.

Principle:

C-reactive protein slide test for determination of C-reactive protein level is based on priniciple of agglutination. The test specimen (serum) is mixed with C-reactive protein latex reagent and allowed to react. If C-reactive protein concentration is greater than 6mg/litre then a visible agglutination is observed.

There are two methods: -

- Qualitative method
- Semiquantitative method

We used both methods. First we used qualitative method to know whether C-reactive protein level was more than 6mg/litre or less than 6mg/litre. If C-reactive protein level in serum was more than 6mg /litre than we used semiquantitative method to know the elevated level of C-reactive protein level in serum.

TEST PROCEDURE :- (For Qualitative measurement)

1. Pipette one drop of serum on to the identified ring of the test glass slide using the disposible pipette provided along with kit.
2. Mix Latex reagent by gentle shaking of the vial. Add one drop of C-reactive protein latex reagent to the drop of test serum on the slide.
3. Using a mixing stick, mix the serum and the latex reagent uniformly over the entire circle.
4. Immediately note the time. Rock the slide gently back and froth. Observing for agglutination macroscopically within two minutes.

TEST PROCEDURE :- (For Semiquantitative measurement)

1. Using isotonic saline prepare serial dilution of the serum sample from 1:2 to 1:64.
2. Pipette 40 microlitre of the diluted serum sample was put on each reaction circle with the pipette which was provided with kit.

Claus et al (1976) observed that C-reactive protein in the neonates is endogenously produced.

C-reactive protein has been studied as a tool for the early diagnosis of neonatal infections (Sabel and Hanson 1974) and was found to be more reliable test as a diagnostic parameter when compared with total leucocyte count and band cell count (Sabel and Wadsworth, 1979).

According to Sabel and Hanson (1974) high C-reactive protein values during first few hours after cinical symptoms had appeared. This suggested that C-reactive protein was sufficiently rapid and specific to serve as a definite and in the early diagnosis of septicemia.

MONITORING AFTER DELIVERY: Parameters studied were - maternal pulse rate, maternal temperature, total lecocyte count of both mother and newborn, differential leucocyte count of both mother and newborn, C-reactive protein determination of both mother and newborn and presence of polymorphs in the gastric aspirate of the newborn (i.e. more than 20 polymorphs/HPF).

Scanlonj (1972) found that leucocytes in the infant stomach are of maternal origin and the bacteria observed on stained smears are probably carried from the nasopharynx to the stomach upon intubation. Gastric aspirates cellularity may be used as screening method for newborn who are likely to develop infection.

Ramos and Stern (1969) were of the opinion that examination of gastric aspirate for polymorphs. While indicating numerically greater number of suspected than subsequently proved infection can nevertheless be used as a rapid, simple and effective means of identifying the infant at risk.

Kumari et al (1983) found gastric aspirate polymorphs a useful side laboratory diagnostic test .The incidence of septicemia bearing a direct association to the number of gastric polymorphs /high power field (i.e. >20 polymorphs/HPF).

All placenta and amniotic membranes were histologically evaluated for evidence of inflammation and/or infection. The criteria that were used to establish a diagnosis of histologic chorioamnionitis were: -

1. Polymorphonuclear leucocyte infiltration of the extra placental membranes.
2. An accumulation of polymorphs in the intervillous space immediately below the chorionic plate.
3. Leucocyte infiltration of chorionic plate.
4. Angitis of umbilical vessels.

MONITORING 6 WEEKS AFTER DELIVERY : Parameters for follow up were - maternal pulse rate, maternal temperature, total leucocyte count of both mother and newborn, differential leucocyte count of both mother and newborn and C-reactive protein determination of both mother and newborn.

After delivery

(a) Cord blood was taken for C-reactive protein of newborn.

(b) Placenta and membranes were sent for histopathological examination (in all possible cases).

(c) Gastric aspirate of newborn for polymorph.

(d) Mother and newborn were followed up for evidence of sepsis.

INVESTIGATIONS:

A. Maternal investigation:

1. Routine blood examination –
   Hb%, TLC, DLC, ESR
2. Maternal C-reactive protein value by using " Latex agglutination method "
3. Maternal cervical swab culture.

B. Investigation of newborn:

Routine blood examination – Hb%, TLC, DLC.

OBSERVATION

OBSERVATION

Present study was conducted in the Department of Obstetrics and Gynaecology in collaboration with Department of Biochemistry in M.L.B. Hospital, M.L.B. Medical College Jhansi since July 2002 to August 2003.

One hundred antenatal cases were selected from out patient department and in-door patient department of obstetrics and gynaecology of M.L.B. Medical College Jhansi. The following samples were analysed.

1. **CASE GROUP**: sixty cases of confirmed premature rupture of membranes before 35 weeks of gestation were studied in this group. Any associated medical illness including infections, rheumatoid arthritis or systemic lupus erythematosus were excluded from the study.

2. **CONTROL GROUP:** forty cases of normal pregnancy were taken as controls.

TABLE - 1

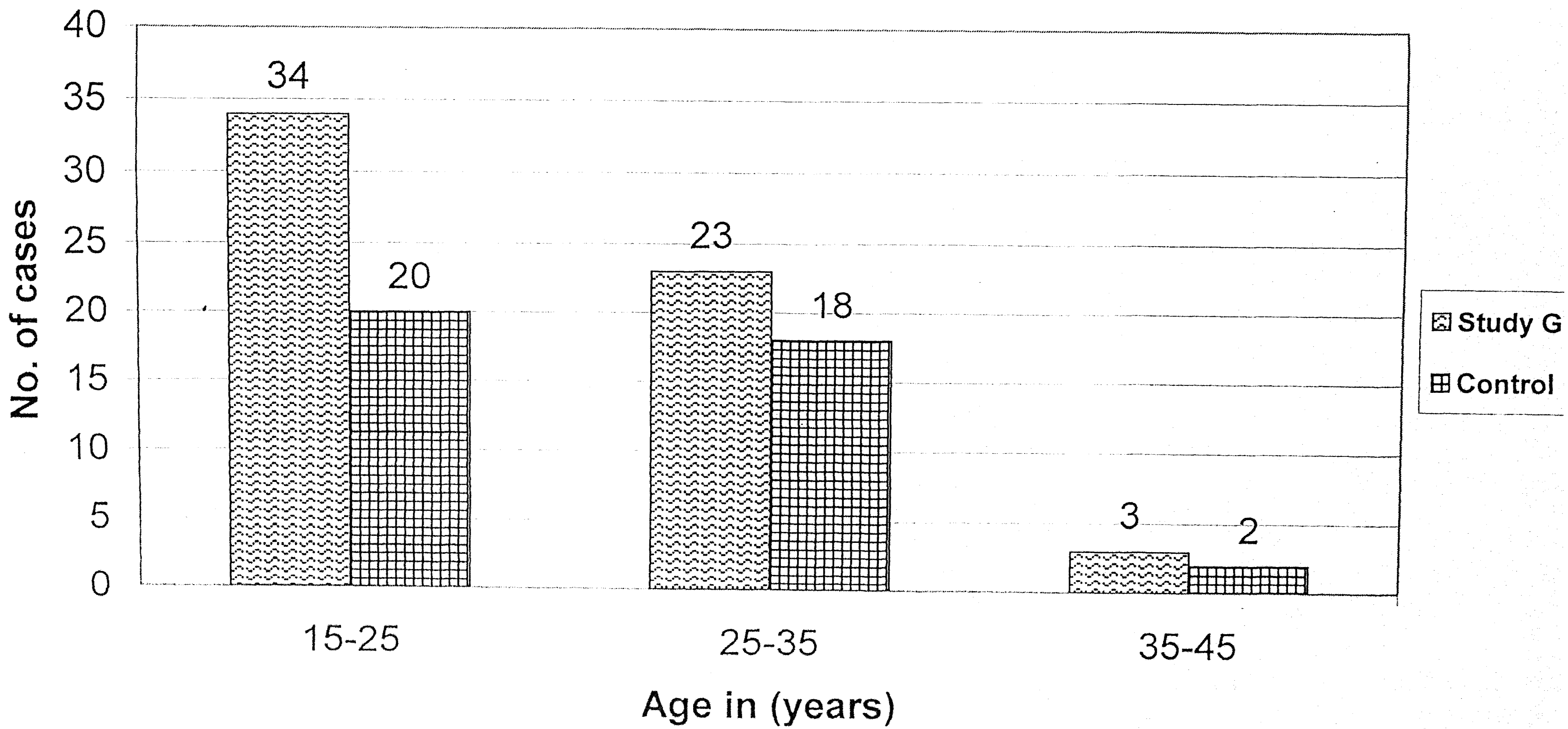
AGEWISE DISTRIBUTION OF CASES IN THE CONTROL AND STUDY GROUP

Age (years)	Study group No.	Study group %	Control group No.	Control group %
15-25	34	56.6	20	50.0
25-35	23	38.4	18	45.0
35-45	3	5.0	2	5.0
Total number	60	100	40	100
Mean ± SD	25.15 ± 5.1		23.9 ± 4.41	

Table-1 and figure-1 has showed highest incidence of premature rupture of membranes in age group between 15-25 years (56.6%) followed by 25-35 years age group (38.4%) and least incidence of premature rupture of membranes in 35-45 years age group (5%).

The study group had higher mean age 25.15±5.1 years than control group 23.9 ± 4.41 years.

AGE WISE DISTRIBUTION OF CASES IN THE CONTROL AND STUDY GROUP



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The study group had higher mean age 25.15±5.1 years than control group 23.9 ± 4.41 years.

SHOWS THE RELATIONSHIP OF THE PREMATURE RUPTURE OF MEMBRANES WITH SOCIO-ECONOMIC STATUS OF PATIENTS

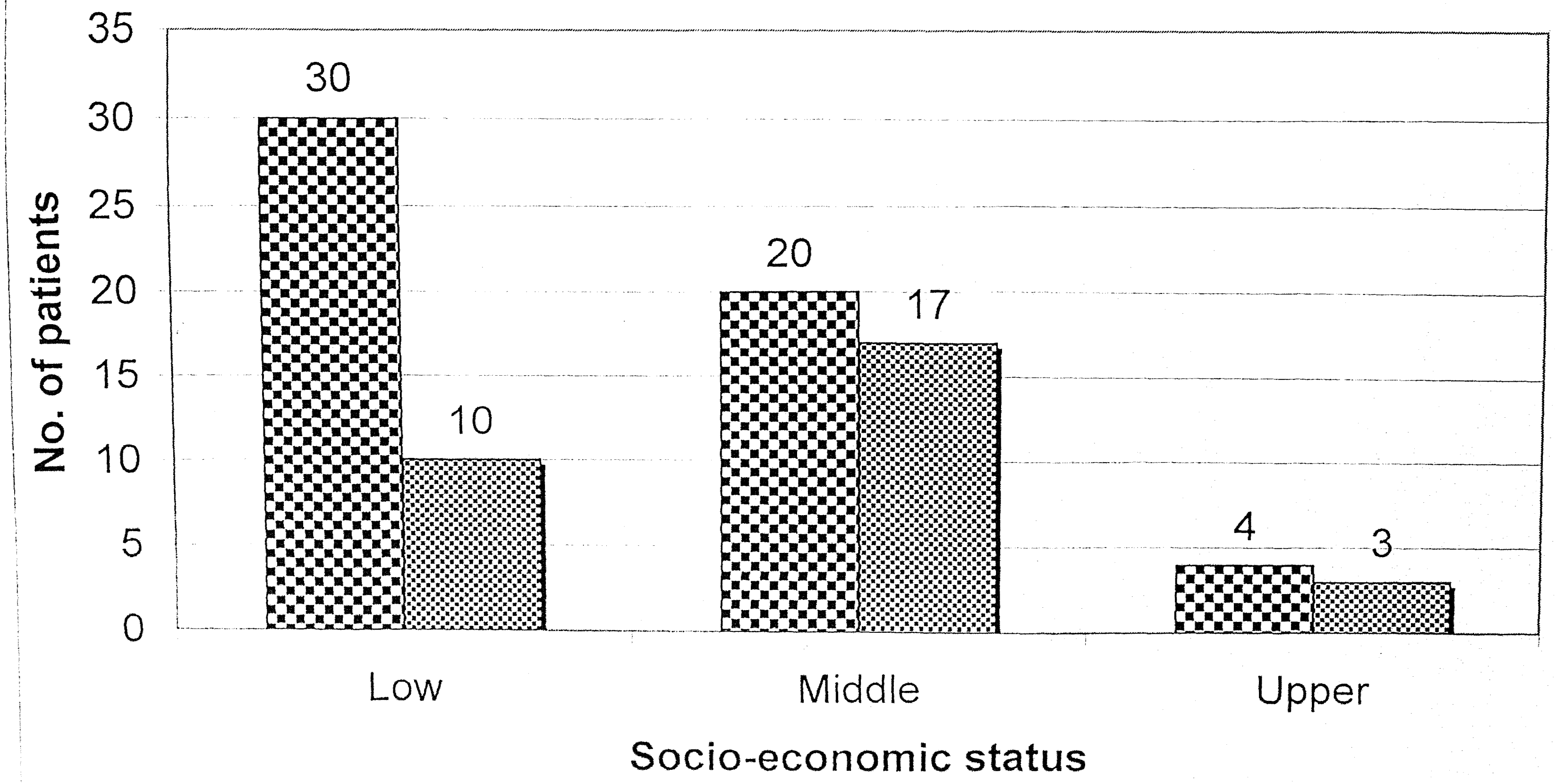


TABLE – 2

SHOWS THE RELATIONSHIP OF PREMATURE RUPTURE OF MEMBRANES WITH SOCIOECONOMIC STATUS OF PATIENTS

Socioeconomic status	Study group		Control group	
	No.	%	No.	%
Low	36	60.0	10	25.0
Middle	20	33.4	17	42.5
Upper	4	6.6	13	32.5
Total number	60	100	40	100

A high incidence of premature rupture of membranes is noted in low socio-economic group 60% as compared to only 6.6% in upper socioeconomic group (Table-2 and Figure-2).

DISTRIBUTION OF CASES ACCORDING TO GRAVIDITY

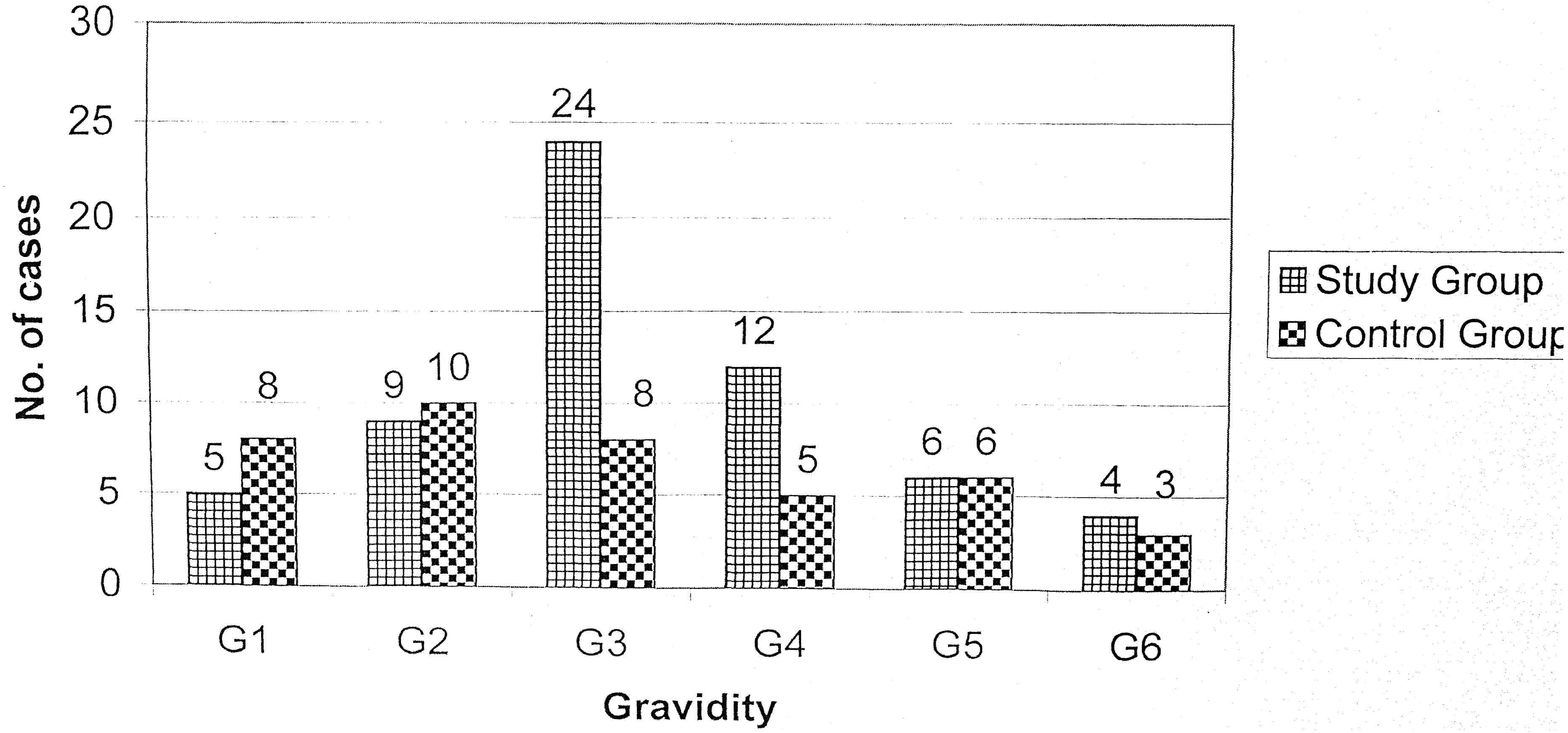


TABLE – 3

DISTRIBUTION OF CASES ACCORDING TO GRAVIDITY

Gravidity	Study group		Control group	
	No.	%	No.	%
G1	5	8.3	8	20.0
G2	9	15.0	10	25.0
G3	24	40.0	8	20.0
G4	12	20.0	5	12.5
G5	6	10.0	6	15.0
G6	4	6.6	3	7.5
Total number	60	100	40	100

Table-3 and Figure-3 shows the distribution of cases according to gravidity. In the study group (i.e. cases with preterm premature rupture of membranes) maximum number of cases were third gravida (40%) followed by forth gravida (20%).

RELATIONSHIP OF PREMATURE RUPTURE OF MEMBRANE WITH GESTATIONAL AGE

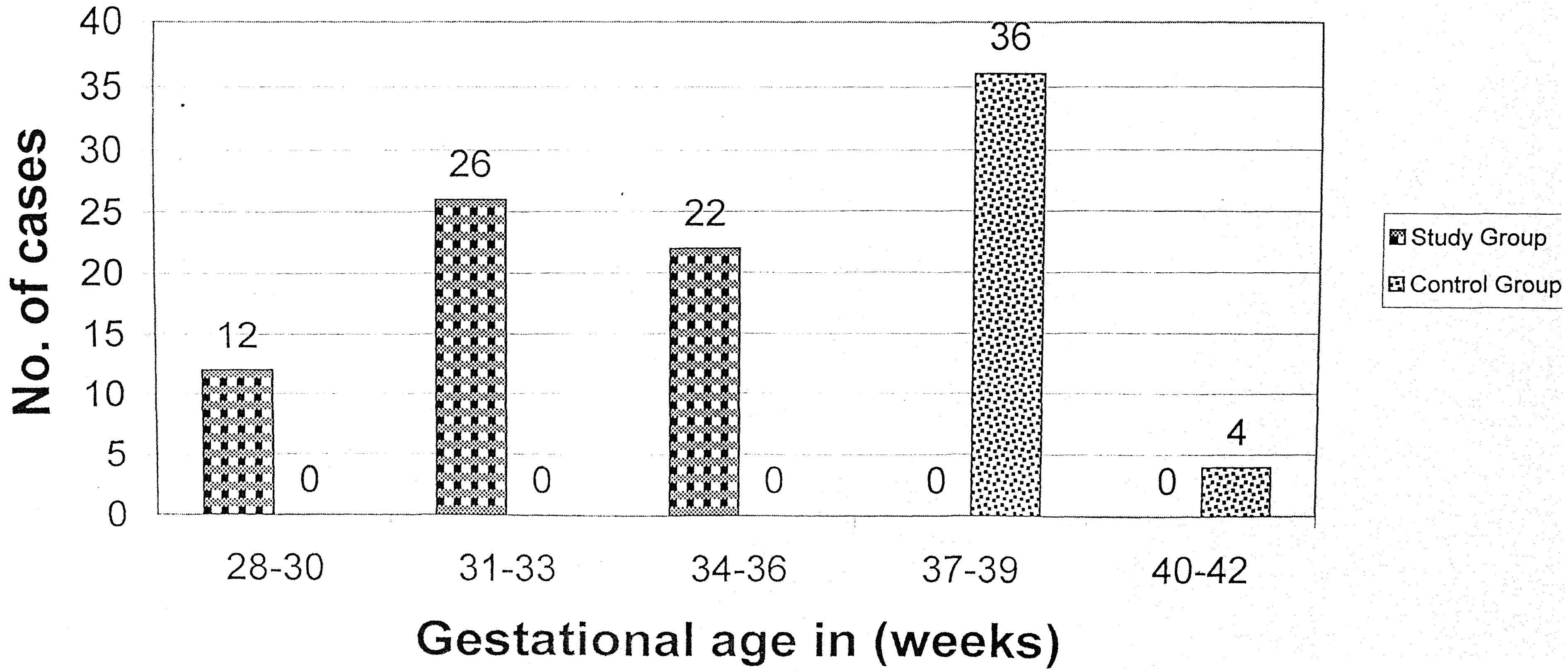


TABLE – 4

RELATIONSHIP OF PREMATURE RUPTURE OF MEMBRANES WITH GESTATIONAL AGE

Gestational age (in weeks)	Study group		Control group	
	No.	%	No.	%
28-30	12	20	-	-
31-33	26	43.33	-	-
34-36	22	36.67	-	-
37-39	-	-	36	90.0
39-41	-	-	04	10.0
Total number	60		40	

Most common gestational age for premature rupture of membranes in our study was 31-33 weeks (43.33%), followed by 34-36 weeks (36.67%) and the least incidence of premature rupture of membranes was in 28-30 weeks of gestational age group cases (Table-4 and Figure-4).

DISTRIBUTION OF CASES ACCORDING TO DURATION OF PREMATURE RUPTURE OF MEMBRANES

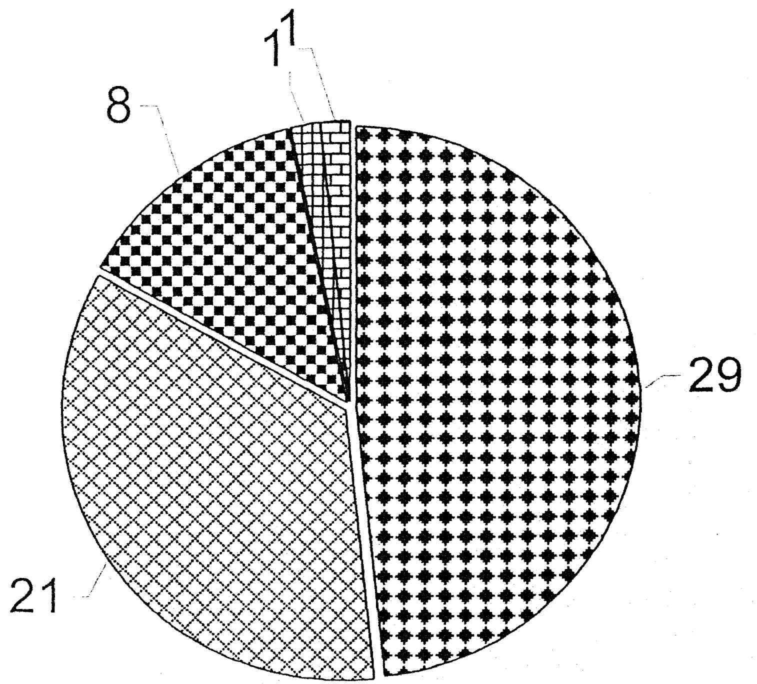


TABLE – 5

DISTRIBUTION OF CASES ACCORDING TO DURATION OF PREMATURE RUPTURE OF MEMBRANES

Duration (in hours)	Study group	
	No.	%
5-145	29	48.3
145-290	21	35.0
290-435	8	13.3
435-580	1	1.7
580-725	1	1.7
Total number	**60**	**100**

Table-5 and figure-5 shows duration of premature rupture of membranes. The highest number of patients (48.3%) had duration of premature rupture of membranes between 5-145 hours followed by 145-290 hours (35.0%) and 290-435 hours (13.3%). Least number of patients (1.7%) had duration of premature rupture of membranes between 435-580 hours & 580-725 hours. Mean duration of premature rupture of membranes was 169.06 hours.

DISTRIBUTION OF CASES ACCORDING TO MODE OF DELIVERY

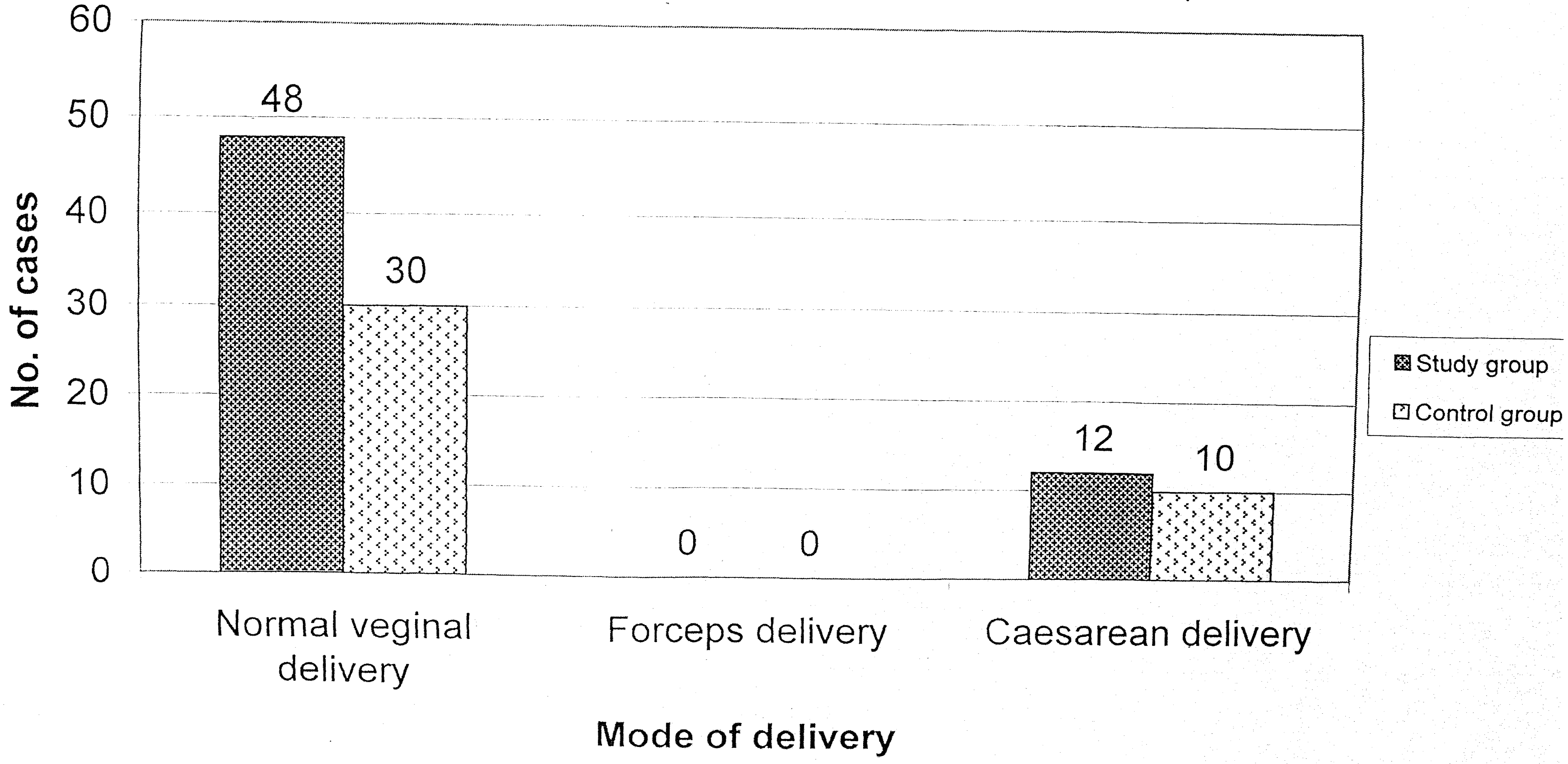


TABLE – 6

DISTRRIBUTION OF CASES ACCORDING TO MODE OF DELIVERY

Mode of delivery	Study group		Control group	
	No.	%	No.	%
Normal vaginal delivery	48	80	30	75
Forceps delivery	-	-	-	-
Caesarean section	12	20	10	25
Total	60	100	40	100

The number of normal vaginal deliveries and caesarean section rate were comparable in normal pregnant female and cases of preterm premature rupture of membranes in table 6 & figure-6.

Percentage of normal vaginal delivery in our study group was 80% while percentage of caesarean section was only 20%.

In control group percentage of normal vaginal delivery was 75% while percentage of caesarean section was 25%.

DISTRIBUTION OF CASES ACCORTING TO FOETALOUTCOME

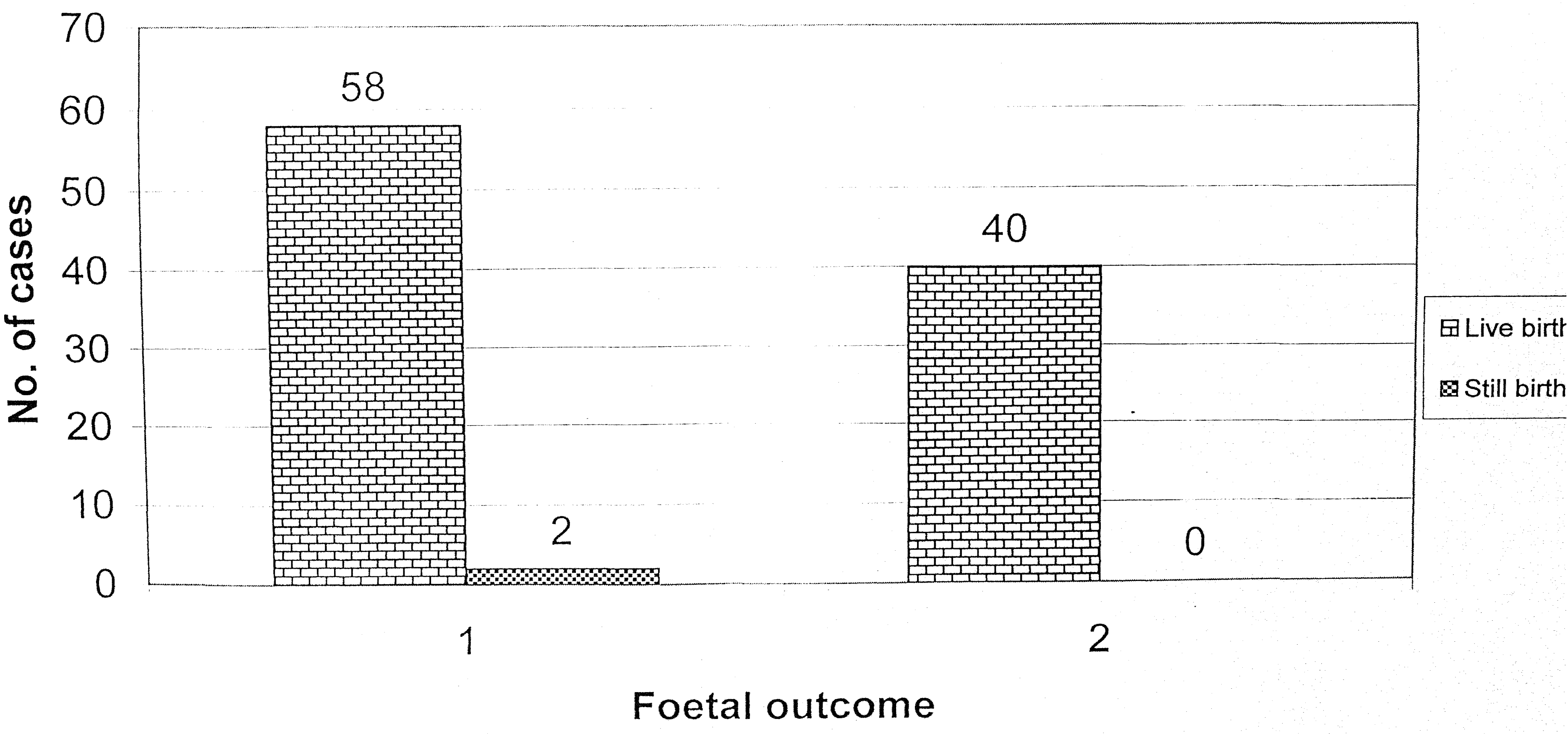


TABLE – 7

DISTRRIBUTION OF CASES ACCORDING TO FOETAL OUTCOME

Foetal outcome	Study group		Control group	
	No.	%	No.	%
Live birth	58	96.7	40	100
Still birth	02	3.3	-	-
Total no.	**60**	**100**	**40**	**100**

In study group total number of infants were 60 and percentage of live birth was 96.7% while only two babies were still born (3.3%)

DISTRIBUTION OF CASES ACCORDING TO THE SEX OF NEWBORN

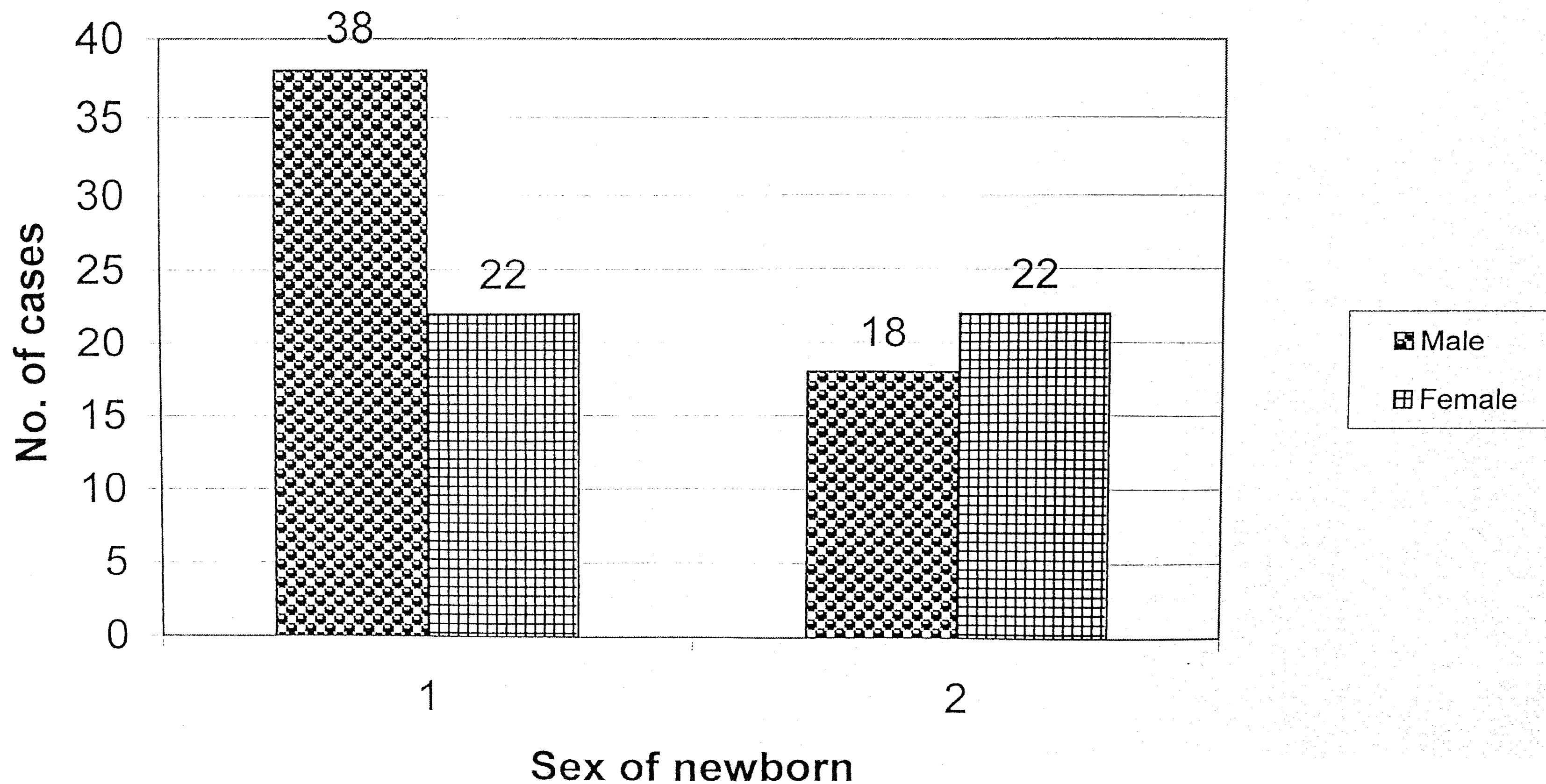


TABLE – 8

DISTRRIBUTION OF CASES ACCORDING TO THE SEX OF NEWBORN

Sex of newborn	Study group		Control group	
	No.	%	No.	%
Male	38	63.4	18	45
Female	22	36.6	22	55
Total	60	100	40	100

Percentage of male newborns was 63.8% and percentage of female newborns was 36.67% in our study group.

In control group percentage of male newborns was 45% and percentage of female newborns was 55%, so percentage of male newborns was more in study group in comparison to control group.

TABLE – 9

MEAN C-REACTIVE PROTEIN LEVELS FOR VARIOUS DIAGNOSTIC GROUPS IN RELATION TO CLINICAL CHORIOAMNIONITIS

Diagnostic groups	Patients in study group		Mean C-reactive protein level in study group
	No.	%	
1. Clinical chorioamnionitis with elevated C-reactive protein level	11	18.33	42.5mg/litre
2. Elevated C-reactive protein level with no clinical chorio-amnionitis	21	35.00	20.1mg/litre
3. Clinical chorioamnionitis with normal C-reactive protein level	01	1.67	<6mg/litre
4. No clinical chorioamnio-nitis with normal C-reactive protein level	27	45.00	<6mg/litre
Total Number	**60**	**100%**	

Among 60 patients included in study group, 11 (18.33%) had clinical chorio-amnionitis with elevated C-reactive protein levels. They had mean C-reactive protein level of 42.5mg/litre. One patient had features of clinical chorio-amnionitis with normal C-reactive protein level i.e. only 1 false positive case. 27 patients in study

group (i.e.45%) had no evidence of clinical chorioamnionitis and their c-reactive protein levels were also in normal range. 21 patients had elevated C-reactive protein level but with no clinical chorioamnionitis. These patients later on found to have histological chorioamnionitis.

TABLE – 10

MEAN C-REACTIVE PROTEIN LEVELS FOR VARIOUS DIAGNOSTIC GROUPS IN RELATION TO HISTOPATOLOGICAL CHORIOAMNIONITIS

Diagnostic groups	Patients in study group		Mean C-reactive protein level in study group
	No.	%	
1. Pathological chorio-amnionitis with elevated C-reactive protein level	31	51.6	26.08mg/litre
2. Elevated C-reactive protein level with no pathological chorio-amnionitis	1	1.6	18mg/litre
3. Pathological chorio-amnionitis with normal C-reactive protein level	3	5.0	<6mg/litre
4. No pathological chorio-amnionitis with normal C-reactive protein level	25	41.8	<6mg/litre
Total	**60**	**100**	

34 patients out of 60 patients with premature rupture of membranes showed histopathological chorioamnionitis on their placental examination. 31 patients, out of these 34, had elevated CRP

values. The mean C-reactive protein level was 26.08mg/litre. Three patients (5%) had pathological chorioamnionitis with normal C-reactive protein level i.e. false negative cases.

Only one patient (1.65%) had elevated C-reactive protein level with no pathological chorioamnionitis, so there was only one false positive case.

TABLE - 11

COMPARISON OF C-REACTIVE PROTEIN DETERMINATION AND OTHER TESTS IN THE IDENTIFICATION OF CLINICALLY DIAGNOSED CHORIOAMNIONITIS

Test	Without Clinical Chorioamnionitis		With Clinical Chorioamnionitis		Sensitivity (%)	Specificity (%)	Positive Predictive Value (%)	Negative Predictive Value (%)
	Test Normal	Test Abnormal	Test Normal	Test Abnormal				
C-reactive protein (>6mg/l)	27	21	1	11	91.6	58.3	35.48	96.5
Maternal temperature	46	2	3	9	75	91.6	69.2	93.6
WBC count (>12000/cumm)	42	6	5	7	58.3	87.5	58.81	89.36
DLC count	44	4	8	4	50	91.6	60	88
Fetal heart rate (>160/min)	40	8	8	4	33	81.2	30.7	82.6
ESR (>60 mm/hr)	48	0	2	10	83.33	100	100	96

In Table-11 we compared C-reactive protein levels with other laboratory tests as indicators of infection (e.g. WBC count, DLC count, ESR, maternal temperature, fetal heart rate). We found C-reactive protein level to be more sensitive (91.6%) but less specific (58.3%) in identifying clinical chorioamnionitis. C-reactive protein was elevated in all cases of clinical chorioamnionitis. C-reactive protein determination was the most sensitive test (91.6%) followed by ESR (83.33%) and presence of fever (75%). Least sensitive were fetal tachycardia (33%). But when specificity was considered C-reactive protein was not specific as other tests. As shown in Table-11 most specific was ESR (100%) followed by presence of fever (91.6%) & DLC count (91.6%).

Thus C-reactive protein was highly sensitive but less specific in diagnosing clinical chorioamnionitis. It was raised at least 48 hours before the onset of signs and symptoms of clinical chorioamnionitis.

TABLE - 12

COMPARISON OF C-REACTIVE PROTEIN DETERMINATION AND OTHER TESTS IN THE IDENTIFICATION OF HISTOPATHOLOGICALLY DIAGNOSED CHORIOAMNIONITIS

Test	Without histological Chorioamnionitis		With histological Chorioamnionitis		Sensitivity (%)	Specificity (%)	Positive Predictive Value (%)	Negative Predictive Value (%)
	Test Normal	Test Abnormal	Test Normal	Test Abnormal				
C-reactive protein (>6mg/l)	25	1	3	31	91.16	96.1	96.8	89.28
Maternal temperatur	25	1	22	10	29.41	96.15	90.90	51.02
WBC count (>12000/cumm)	23	3	22	10	29.41	88.46	76.92	48.93
DLC count	23	2	28	06	17.64	92.3	75	46.15
Fetal heart rate (>160/min)	22	4	22	09	26.47	84.61	69.23	46.80
ESR (>60 mm/hr)	26	0	24	10	29.41	100	100	52

Table-12 shows comparison of sensitivity and specificity of different tests in identification of hisotpahtological chorioamnionitis. We found that C-reactive protein level was very sensitive (91.16%) and specific (96.1%) in identification of hisotpahtological chorioamnionitis. ESR, though, very specific (100%) but very less sensitive (29.41%) in identification of hisotpahtological chorioamnionitis.

34 patients out of 60 patients (56.6%) in study group showed moderate to severe inflammation on the histological examination of their placenta. Only one patient (1.66%) with no evidence of histopathological chorioamnionitis had mildly raised C-reactive protein value (12mg/litre) i.e. false positive case. Mild leucocytic infiltration was found in both study and control group and difference was not statistically significant. Three patients (5%) with normal C-reactive protein values had histopathological chorioamnionitis i.e. false negative cases.

Least sensitive test in identification of hisotpahtological chorioamnionitis was DLC count (17.64%) followed by foetal tachycardia (26.47%).

TABLE-13

C-REACTIVE PROTEIN LEVELS CORRELATED WITH PROLONGATION OF GESTATION

C-reactive protein	N	Mean hours to delivery
Negative	28	264.10± 26.2
Positive	32	85.87± 7.7
Total	60	

Table-13 shows the correlation of C-reactive protein with the prolongation of gestation. The mean hours to delivery for the patients with negative C-reactive protein value were 264.1±26.2 hours versus 85.87±7.7 hours for patients with positive C-reactive protein value ($p<.01$).

C-REACTIVE PROTEIN CORROLATED WITH RESULT OF TOCOLYSIS

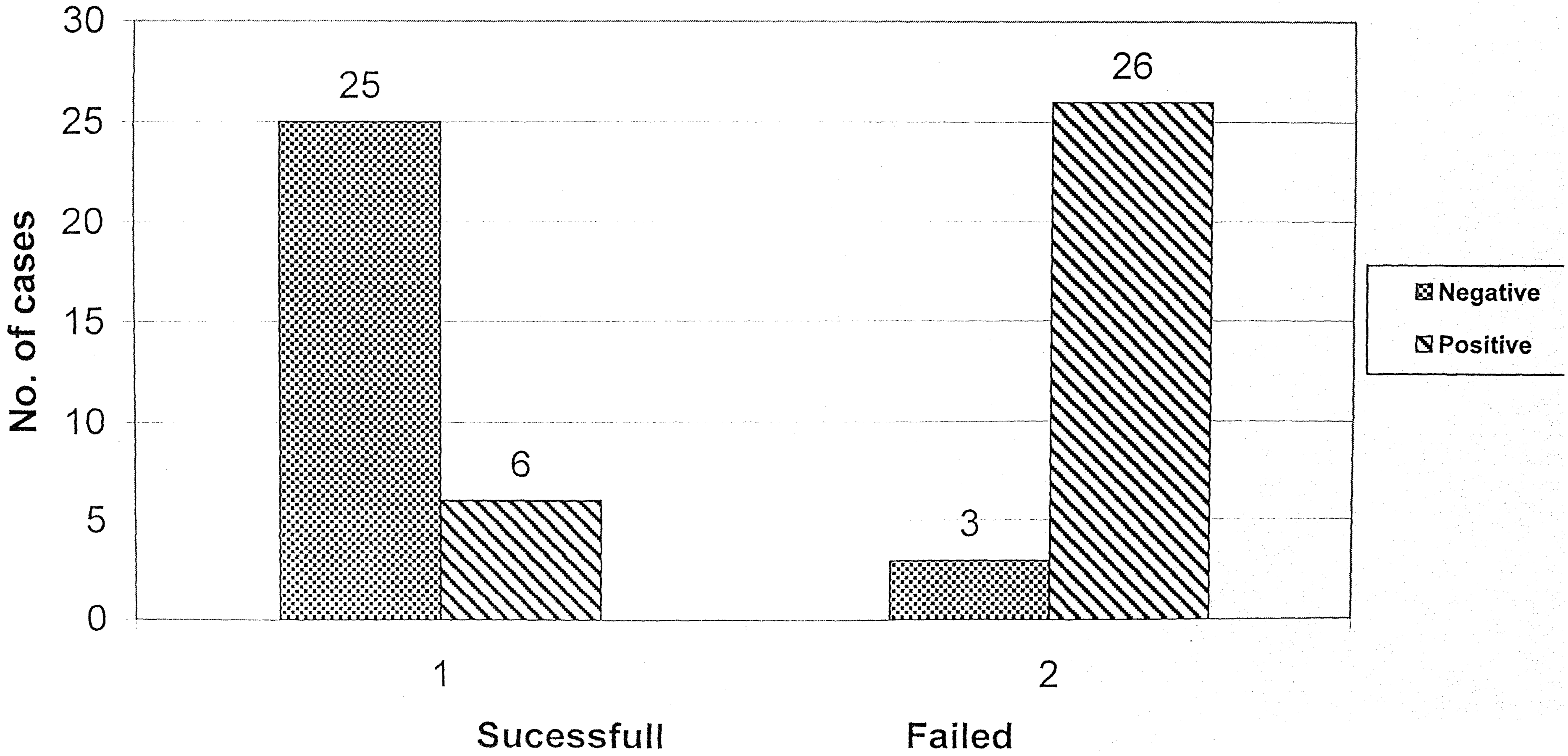


TABLE – 14

C-REACTIVE PROTEIN CORRELATED WITH RESULTS OF TOCOLYSIS

C-reactive protein	N	Successful		Failed	
		N	%	N	%
Negative	28	25	89.3	3	10.7
Positive	32	6	18.7	26	81.3

Table 14 shows the correlation of C-reactive protein value with the results of tocolysis. Tocolysis was successful in prolonging gestation for 1week in 25 of the 28 patients (89.3%) with a negative C-reactive protein value while tocolysis was successful only in 6 of the 32 patients (18.7%) with positive C-reactive protein value. Tocolysis was significantly more successful in the patients with negative C-reactive protein value.

TABLE-15

MATERNAL MORBIDITY IN PREMATURE RUPTURE OF MEMBRANES

Causes of maternal morbidity	Study group (n=60)			
	No.	%	No. of cases with positive cervical swab	No. of cases with raised CRP level (>6mg/litre)
1. Puerperal pyrexia	11	18.3	5	5
a. Puerperal sepsis	05	08.3	5	5
b. Miscellaneous	06	10.0	-	-
1. Malaria	03	05.0	-	-
2. Idiopathic	03	05.0	-	-
2. Postpartum hemorrhage	00	00.0	-	-

Table 15 is showing maternal morbidity in premature rupture membranes.

Total maternal morbidity rate was 18.3 %. The major cause of maternal morbidity was chorioamniotis, which later on developed into puerperal sepsis (due to late coming of the patients in the hospital).

In our study out of eleven patients of puerperal pyrexia five were suffering from puerperal sepsis (8.3%). All the five had positive cervical swab culture and elevated C-reactive protein level.

Out of five, four had positive cervical swab culture for Esch. Coli and had sensitively with antibodies like Amikacin, Cefotaxin, Netlimycin, gentamycin. The remaining one had positive cervical swab culture for Klebsiella species and had sensitivity with antibodies like Ceftriaxone, cefotaxim, Cefoperazone, Netlimycin, Amikacin.

All the five patients were treated with proper antibiotics and responded well.

Remaining six patients had normal C-reactive protein value. Out of six, three were suffering from malaria (Plasmodium vivax). and were treated with antimalarial drugs while the other three patients with idiopathic fever became afebrile on second postnatal day without any treatment.

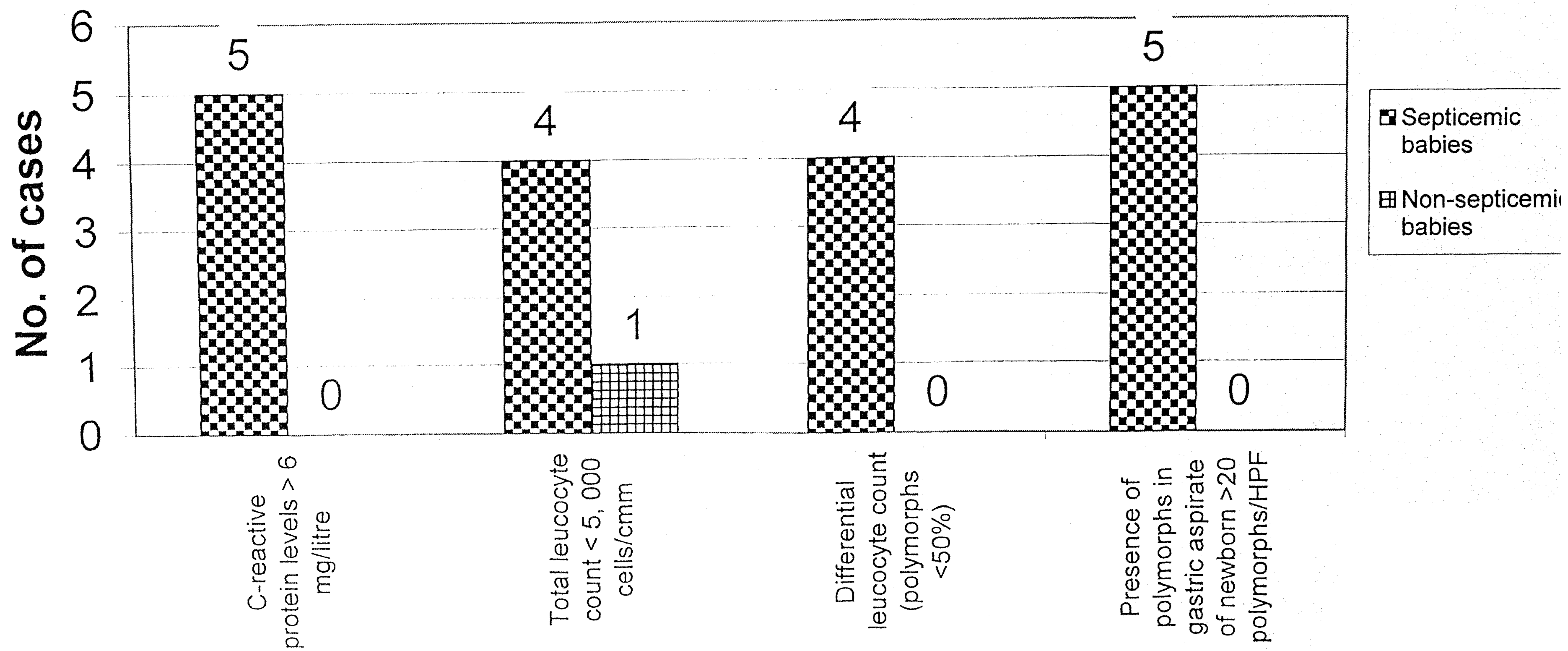
TABLE – 16

SHOWING CORRELATION OF ELEVATED C-REACTIVE PROTEIN LEVEL AND NEONATAL MORBIDITY IN PREMATURE RUPTURE OF MEMBRANES

Cause of neonatal morbidity	Study group (n = 58)			
	No.	Average value of C-reactive protein in mg/litre	No. of cases showing C-reactive protein value above 6mg/litre	% of cases showing C-reactive value above 6 mg/litre
1. Neonatal sepsis	5	20.8	5	8.62%
2. Neonatal hyperbilirubin-aemia	9	<6	-	-
3. Neonatal respiratory distress syndrome	3	<6	-	-
4. Neonatal intraventricular haemorrhage	1	<6	-	-

Table – 16 is showing correlation of elevated C-reactive protein level in neonatal blood and neonatal morbidity in premature rupture membranes.

COMPARISION OF C-REACTIVE PROETIN LEVELS ACCURACY WITH OTHER TESTS IN DIAGNOSING NEONATAL SEPSIS IN OTHER GROUP



C-reactive protein was raised (more than 6 mg/litre) in all the 5 septicaemic babies (8.6%). It was not raised (less than 6 mg/litre) in other causes of neonatal morbidity (like neonate hyperbilirubinaemia, neonatal respiratory distress syndrome and neonatal intraventricular haemorrhage).

Average value of C-reactive protein in septicemic babies was 20.8 mg/litre.

TABLE – 17

COMPARISON OF C-REACTIVE PROTEIN LEVELS ACCURACY WITH OTHER TESTS IN DIAGNOSING NEONATAL SEPSIS IN STUDY GROUP

Tests	Septicemic babies (n = 5)		Non-septicemic babies (n =58)	
	No.	%	No.	%
1. C-reactive protein levels > 6 mg/litre	5	100	-	-
2. Total leucocyte count < 5, 000 cells/cmm	4	80	1	1.8
3. Differential leucocyte count (polymorphs <50%)	4	80	-	-
4. Presence of polymorphs in gastric aspirate of newborn >20 polymorphs/HPF	5	100	-	-

In table – 17 we have compared neonatal C-reactive protein levels with other tests as indicator of neonatal sepsis (e.g total leucocyte count, differential leucocyte count and presence of polymorphs in gastric aspirate of newborn).

We found C-reactive protein level was raised (>6 mg/litre) in all the 5 septicemic babies. Thus accuracy of C-reactive protein level in neonatal blood is 100%.

Leucopenia (<5,000 cells/cmm) was present in 4 babies out of 5 septicemic babies and had an accuracy of 80%.

Percentage of polymorphs less than 50% in 4 out of 5 septicemic babies and had an accuracy of 80%.

Presence of polymorphs more than 20/HPF in gastric aspirate of newborns was present in all the 10 septicemic babies. Thus had an accuracy of 100%.

In non-septicemic babies (n=53) C-reactive protein levels was not raised and did not have polymorphs in gastric aspirate of newborn. Only one nonsepticemic babies had leucopenia.

Thus we found C-reactive protein level was raised in the 5 septicemic babies. Thus showed 100% accuracy.

PERINATAL MORBIDITY IN PREMATURE RUPTURE OF MEMBRANE

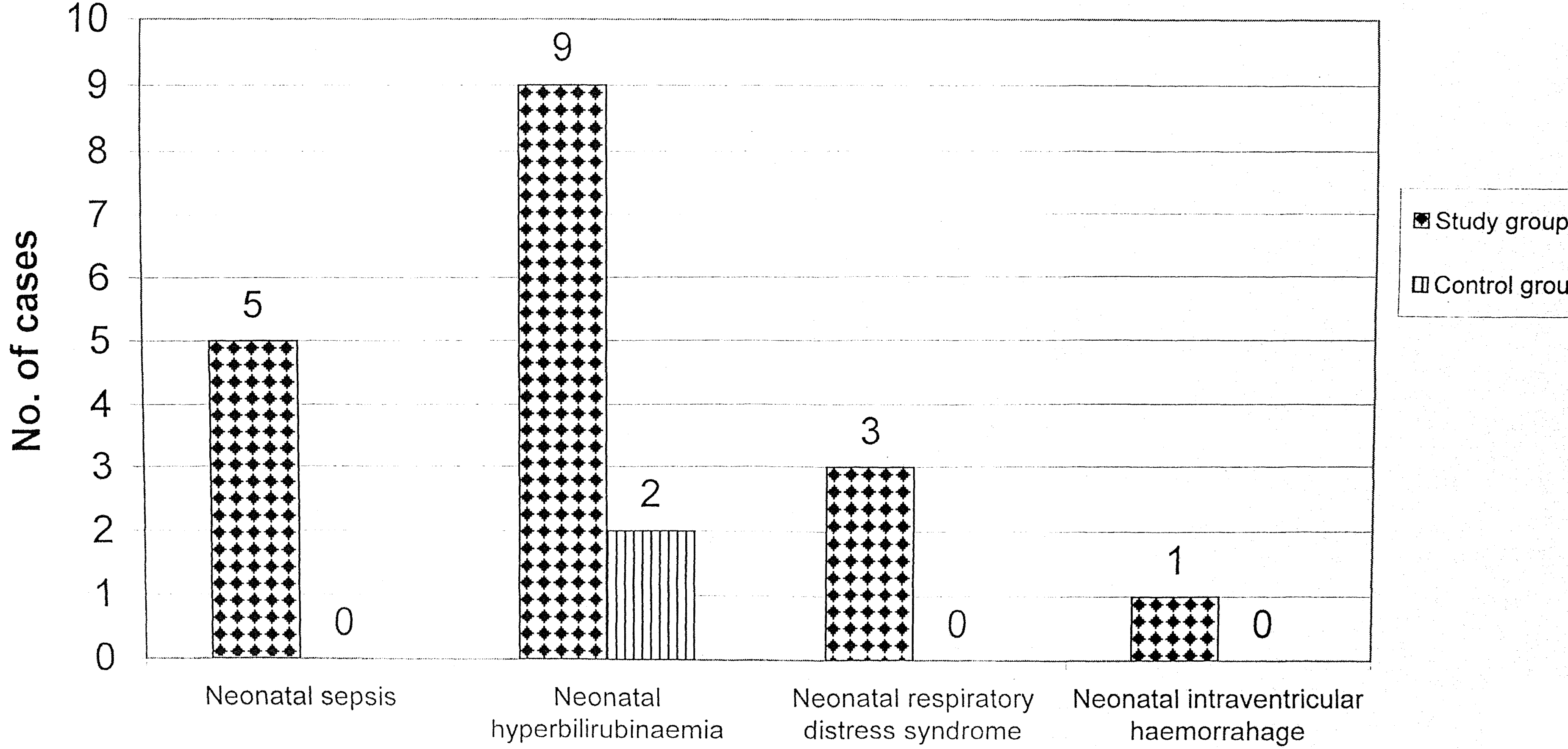


TABLE – 18

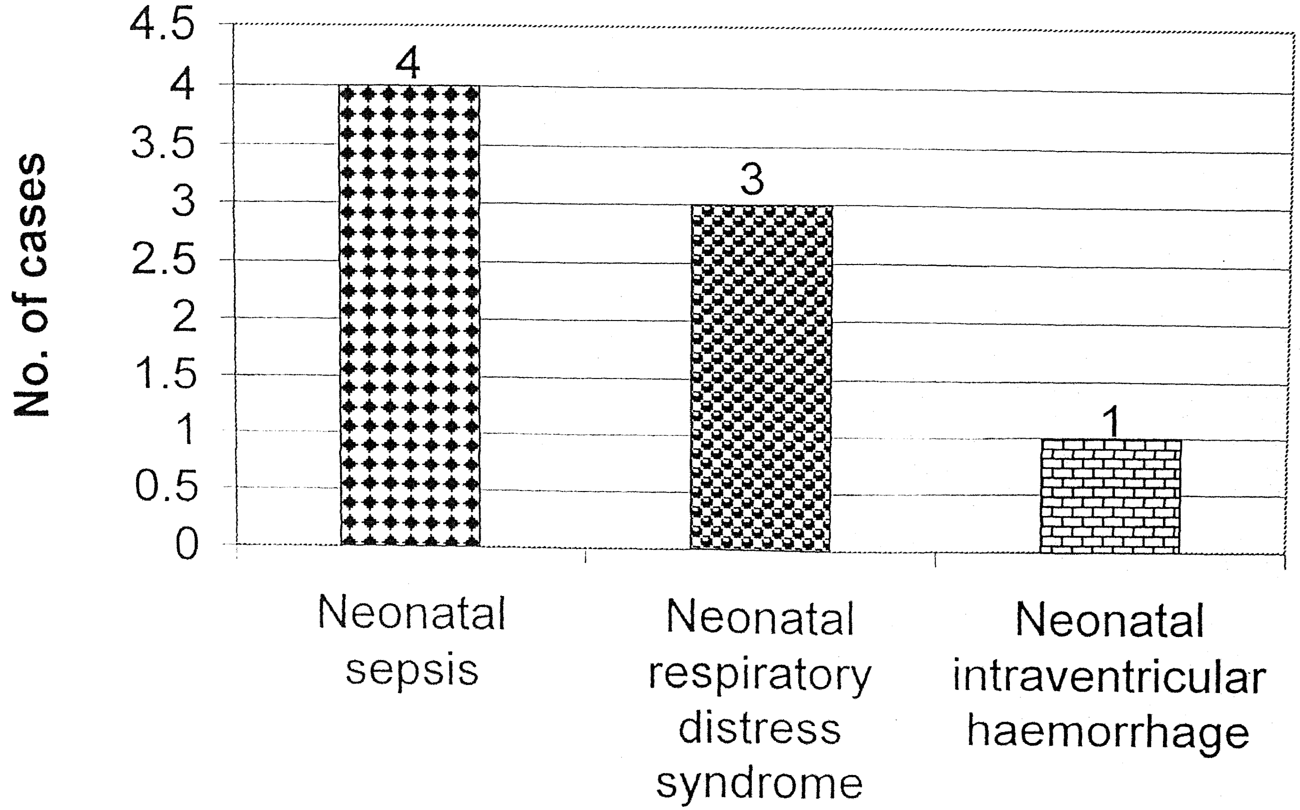
PERINATAL MORBIDITY IN PREMATURE RUPTURE OF MEMBRANES

Causes of perinatal morbidity in premature rupture of membrane	Study group (n=58)		Control group (n=40)	
	No.	%	No.	%
1. Neonatal sepsis.	5	8.63	-	-
2. Neonatal hyperbilirubinaemia.	9	15.51	2	4.0
3. Neonatal respiratory distress syndrome.	3	5.17	-	-
4. Neonatal intraventricular haemorrahage.	1	1.72	-	-
Total	**18**	**Total perinatal morbidity 31.03%**		**Total perinatal morbidity = 4.0%**

Table – 18 have shown total perinatal morbidity rate in premature rupture of membranes. Causes of perinatal morbidity are neonatal sepsis, neonatal hyperbilirubinaemia, neonatal respiratory distress syndrome and intraventricular haemorrhage in study group.

Most common complication of premature rupture of membranes in perinatal morbidity is neonatal hyperbilirubinaemia (15.51%) followed by neonatal sepsis (8.62%) and neonatal respiratory distress

<u>**PERINATAL MORTALITY IN PREMATURE RUPTURE OF MEMBRANE**</u>



syndrome (5.17%). Least common complication of premature rupture of membranes in perinatal morbidity is neonatal intraventricular haemorrhage (1.7%). Total perinatal morbidity in our study group was 31.03%.

TABLE – 19

PERINATAL MORTALITY IN PREMATURE RUPTURE OF MEMBRANES

Causes of perinatal mortality.	Study group (n=58%)		Percentage of total perinatal mortality
	No.	%	
1. Neonatal sepsis.	4	6.89	50.0
2. Neonatal respiratory distress syndrome.	3	5.17	37.48
3. Neonatal intraventricular haemorrhage.	1	1.73	12.51
Total	**8**	**13.79**	

Table – 19 has shown causes of perinatal mortality along with percentage of total perinatal mortality rate due to individual causes.

Total perinatal mortality rate was 13.79%, 50% of which was attributed to sepsis followed by 37.48% to neonatal respiratory distress syndrome and 12.51% to neonatal intraventricular haemorrhage.

DISCUSSION

Expectant management for preterm premature rupture of membranes is now an accepted modality of treatment. Nevertheless, the main clinical concern is still in danger to the mother of acquiring choriamnionitis. Therefore, the approach to expectant management is based on monitoring for signs and symptoms of impending infection. The laboratory indicators, most often used to predict infection, are total leucocyte count, differential leucocyte count & ESR. The tests are, by and large, unreliable because of a very wide range for normal values in pregnancy and additional factors that could affect the result, such as stress or physical activities. Spiking fever, the most reliable clinical symptom, appears when the infection is already established.

C-reactive protein appears to be the most sensitive acute phase protein, rising thousand folds in the initial stages of inflammation. A short half-life of less than 24 hours makes it suitable to serve as a marker for diagnosing an infective process in early stage.

The purpose of this study was to evaluate the usefulness of C-reactive protein determination in the diagnostic process of clinical Chorioamnionitis in patients with preterm premature rupture of membranes.

We have conducted a chronologic comparison with other laboratory tests and clinical signs to establish the sequence for appearance of clinical chorioamnionitis. In our study total, hundred patients were studied. The control group consisted of forty antenatal patients with full term normal pregnancy whereas; study group was comprised of sixty antenatal patients with preterm premature rupture of membranes.

(Any associated medical illness including rheumatoid arthritis or systemic lupus erythematosus was excluded from the study).

Age wise distribution of patients was compared in the study and control group (table-1). Highest incidence of preterm premature rupture of membranes occurred in the cases of 15 - 25 years age group, and mean age of the study group was 25.15 ± 5.1 years which is higher than control group (23.9 ± 4.41 years).

Naeye and Peter (1980) reported that advanced maternal age is an associated factor for preterm premature rupture of membranes.

The mean age of antenatal patients with premature rupture of membranes was 25.2±0.7 years in the study of Y. Romen and R. Artal (1984) which is consistent to our findings.

Socioeconomic status of patients was correlated with premature rupture of membranes (Table-2). A high incidence of preterm premature rupture of membranes was noted in low

socioeconomic group (60%) as compared to upper socioeconomic group (6.6%) in our study. Our findings are in agreement with the findings of Artal et al (1990). They found that defect in the membranes may arise because of poor nutritional status which is significantly influenced by socioeconomic status of the patients.

In the present study maximum number of patients with premature rupture of membranes belonged to 3rd gravida (40.0%) followed by fourth gravida (20.0%).

Keirise MJNC, Rush RW, Anderson ABM, Turnbell AC et al (1978) found that highest risk of preterm premature rupture of membranes occurred in women with two or more previous births and had a 70% chance of the repetition the process.

Y. Romen and R. Artal (1984) and Mohmoud A Ismail, Michael J Zinaman, Richard I. Lowensohn and Atef H. Moawad (1985) also reported maximum number of preterm premature rupture of membranes in third gravida, which is consistent with our findings.

Most common gestational age group for premature rupture of membranes in our study was 31-33 weeks (43.33%). Y. Romen and R. Artal (1984) reported similar results. Their mean gestational age was 30.4 ± 0.4 weeks. Our findings are also similar to Mohmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn, Atef H. Moawad (1985). They found mean gestational age 31 weeks.

In our study group the highest number of patients (48.3%) had duration of premature rupture of membranes between 5-145 hours followed by 145-290 hours (35.0%) and 290-435 hours (13.3%). Least number of patients (1.7%) had duration of premature rupture of membranes between 435-580 hours & 580-725 hours. Mean duration of premature rupture of membranes at the time of delivery was 169.06 hours. Our finding is consistent with the findings of Ismail MA, Zinaman MJ, Lowensohn RI, Moawad AH. and associates (1985). They found mean duration of premature rupture of membranes 150 hours.

In our study the number of normal vaginal deliveries and caesarean section rate were comparable in the control and study group. Percentage of normal vaginal delivery in the study group was 80% while percentage of caesarean section was only 20%. In control group percentage of normal vaginal delivery was 75% while percentage of caesarean section was 25%.

TABLE-20

SHOWING MODE OF DELIVERY IN PREMATURE RUPTURE OF MEMBRANES

Study	Normal vaginal delivery %	Vaccum rotation and extraction %	Forceps vaginal delivery %	Caesarean section %
Mohmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn, and Atef H. Moawed (1985)	47.00	-	40.0	13.00
Anjana Devi and Reddi Rani (1996)	42.30	4.8	07.6	45.20
Our findings	80.0	-	-	20.0

Our findings are parallel to Mohmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn and Atef H. Moawad (1985). In their findings caesarean section rate was 13%, spontaneous vaginal delivery rate was 47%. Anjana Devi and Reddi Rani (1996) found higher incidence of caesarean section (45.20%) as compared to normal vaginal deliveries (42.30%).

In present study total number of newborns deliver were sixty (Table-7). In study group only two babies were stillborn (3.3%). Percentage of live births was 96.7%.

TABLE-21

SHOWING FOETAL OUTCOME IN PREMATURE RUPTURE OF MEMBRANES (STUDY GROUP)

Study	Live birth (%)	Still birth (%)
Mohmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn, and Atef H. Moawed (1985)	98.0	2.00
Anjana Devi and Reddi Rani (1996)	97.1	2.90
Our findings	96.7	3.3%

Our findings are in accordance with Mohmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn, and Atef H. Moawed (1985). Their percentage of live birth was ninety-eight and percentage of stillbirth was two.

Our findings are also similar to Anjana Devi and Reddi Rani (1996). Their percentage of live births was 97.1% and percentage of stillbirth was 2.9%.

In this study percentage of male newborns was 63.80% and percentage of female newborns was 36.67% in the study group. In control group percentage of male newborns was 45% and female newborns was 55%.

TABLE-22

SHOWING PERCENTAGE OF SEX DISTRIBUTION OF FOETUS IN PREMATURE RUPTURE OF MEMBRANES

Study	Male (%)	Female (%)
Mahmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn and Atef H. Moawed (1985)	64.0	36.00
Our study	63.80	36.67

Our findings supported those of Mahmoud A. Ismail, Michael J. Zinaman, Richard I. Lowensohn and Atef H. Moawed (1985). They found percentage of male newborns 64% and female newborns 36%.

The correlation of mean C-reactive protein level in various diagnostic groups for clinical chrioaminiotic has been shown in table-9. There were 4 diagnostic groups. First group had antenatal patients with preterm premature rupture membrane with signs and symptoms of clinical chrioamnionitis with elevated C-reactive protein level. They were only eleven in number (18.33%) and had mean C-reactive protein level 42.5 mg/litre. Second group had antenatal patients with preterm premature rupture of membrane with no signs and symptoms of clinical chorioamnionitis but had elevated level of C-reactive protein value (20.1 mg/litre). The percentage of such patient was 35% (total 21). These patients showed histopathological chorioamnionitis on histological examination of placenta.

Third group had only one antenatal patient with preterm premature rupture of membrane with signs and symptoms of clinical chorioamnionitis but with C-reactive protein value less than 6 mg/litre. This patient didn't show histological chorioamnionitis. This patient latter found to have U.T.I. 4th group had normal CRP level with no clinical chorioamnionitis. The percentage of such patient was 45%. On histological examination also there was no histological chorioamnionitis.

Our findings support the view of Y.Romen and R. Artal by (1984) and Anu Mathur, S.S. Trivedi et al (1992) (table –23). They found that C-reactive protein is a good predictor of infectious morbidity as compared to other indicators. According to them C-reactive protein was elevated in all the cases of clinical chorioamnionitis. The mean value was 31.3 mg/litre in clinical chorioamnionitis. There was only one false positive case with C-reactive protein value 15 mg/litre (i.e. with no signs and symptoms of clinical chorioamnionitis).

TABLE – 23

SHOWING MEAN C-REACTIVE PROTEIN LEVELS FOR VARIOUS DIAGNOSTIC GROUP BY DIFFERENT WORKERS

Study	Diagnostic group	% of patients	CRP in mg/litre
Y.Romen and R. Artal (1984)	Clinical chorioamnionitis with elevated C-reactive protein level.	13.70	46.00
	Elevated C-reactive protein protein level with no clinical chorioamnionitis.	15.70	41.00
	No clinical chorioamnionitis clinical with normal C-reactive protein level.	70.60	8.20
Anu Mathur and S.S. Trivedi et al (1992)	Clinical chorioamnionitis with elevated C-reactive protein level.	6.67	31.16
	Elevated C-reactive protein protein level with no clinical chorioamnionitis.	2.20	15.00
	No clinical chorioamnionitis with normal C-reactive protein level.	91.11	< 6
Our findings	Clinical chorioamnionitis with elevated C-reactive protein level.	18.33	42.5
	Elevated C-reactive protein protein level with no clinical chorioamnionitis.	35.0	20.1
	No clinical chorioamnionitis with normal C-reactive protein level.	45.0	< 6

We compared C-reactive protein with other tests as indicators of infections (e.g. total leucocyte count, different leucocyte count, maternal temperature and foetal heart rate, ESR). C-reactive protein determination was the most sensitive test (91.6%) followed by presence of fever (75%) and total leucocyte count (58.3%). Least sensitive was foetal tachycardia and differential leucocyte count but when specificity was considered C-reactive protein is not as specific as other tests (table-11). Most specific was the ESR (100%) followed by foetal tachycardia (81.2%) & TLC count of 75%. Thus we found that C-reactive protein was highly sensitive (91.6%) but less specific (58.3%) in the identification of clinical chorioamnionitis. Our findings are similar to Harry. F. Farb, Mark Arnesen, Patricia Geistler and G. Eric Knox (1983). They found sensitivity 56% and specificity 73% in diagnosing clinical choriamnionitis. But our findings slightly differ from that of Mark I. Evan, Samir N. Hajj, Lawerence, D. Devoe, Neil S. Angerman and Atef H. Moawad (1980). They found sensitivity 80% and specificity 100% in predicting infections.

Our findings are also in accordance with Y. Romen and R. Artal (1984). According to them C-reactive protein appears to be more accurate in diagnosing clinical chorioamnionitis when compared to total leucocyte count and differential leucocyte count. They found sensitivity of 97% in diagnosing clinical chorioamnionitis. Our findings are similar to Mahmoud A. Ismail, Michel J. Zinaman, Richard I. Lowensohn and Atef H. Moawad (1985). According to them sensitivity of C-reactive protein in diagnosing clinical chorioamnionitis was 82% and specificity was 55%. Our findings are in agreement with the earlier observation

made by Anu Mathur, S.S. Trivedi et al (1992). They found sensitivity 100% and specificity 97.6%.

In our study 56.6% patients in study group had histopathological chorioamnionitis. 91.1% patients with histopathological chorioamnionitis had elevated C-reactive protein level while 8.8% had normal C-reactive protein level. The mean C-reactive protein level was 26.08mg/litre. There was only one (1.65%) false positive case.

We also compared the sensitivity and specificity of different tests in identification of pathological chorioamnionitis. We found that C-reactive protein determination was most specific (96.1%) (Presence of fever also 96.1% specific) followed by DLC count (92.3%), total leucocyte count (80%), & foetal tachycardia (84.61%) ESR though 100% specific but is less sensitive test for identification of histological chorioamnionitis. Thus we found that C-reactive protein determination was less sensitive in identification of histological chorioamnionitis but still most sensitive test (91.16%) followed by total leucocyte count (29.4%), ESR (29.4%) and presence of fever in mother (29.4). Least sensitive test in identification of histological chorioamnionitis was foetal tachycardia (26.4%). Mild leucocyte infiltration was found in both study and control groups and differences were not statistically significant. Sensitivity of C-reactive protein in predicting moderate to severe chorioamnionitis was 91.16% whereas specificity was 96.1%. Our findings resemble with the findings of Mark I. Evan, Samir N. Hajj, Lawerence, D. Devoe, Neil S. Angerman and Atef H. Moawad (1980). They found 100%

accuracy in predicting histological chorioamnionitis by C-reactive protein. According to **Hawrylyshyn P, Bernstein P, Milligan JE, Soldin S, Pollard A, Papsin FR.** (1983) elevated C-reactive protein levels correlated better with pathologic confirmation of chorioamnionitis than with the clinical febrile morbidity. It was raised at least 48 hours before onset of signs and symptoms of clinical chorioamnionitis. White blood cell counts, band neutrophile count, and erythrocyte sedimentation rate were found to be unreliable. C-reactive protein determination was found most reliable with a high degree of sensitivity and specificity. 61.9% had significant chorioamnionitis on histopathology, however, only 16.7% developed clinical signs of chorioamnionitis that is consistent with our findings. (In our study we found that 56.6% cases had histopathological chorioamnionitis while 18.3% had clinical chorioamnionitis). According to Harry. F. Farb, Mark Arnesen, Patricia Geistler and G. Eric Knox (1983) sensitivity of C-reactive protein in identification of histological chorioamnionitis was 80% and specificity was 68%, which is not consistent with our findings. Mahmoud A. Ismail, Michel J. Zinaman, Richard I. Lowensohn and Atef H. Moawad (1985) reported that elevated C-reactive protein levels were correlated well with pathological diagnosis of chorioamnionitis with a sensitivity of 67% and specificity of 81%. Our findings differ from that of Anu Mathur, S.S. Trivedi et al (1992). They found that sensitivity of C-reactive protein in predicting moderate to severe chorioamnionitis was 57.2% whereas specificity was 100%.

In table-13 correlation of C-reactive protein with the prolongation of gestation is shown. The mean hours to delivery for

the patients with negative C-reactive protein value were 264.1 hours while the same was 85.87 hours for patients with positive C-reactive protein value ($p<.01$).

When C-reactive protein levels were correlated with the results of tocolysis we found that tocolysis was successful in prolonging gestation for 1week in 25 of the 28 patients (89.3%) with a negative C-reactive protein value while tocolysis was successful only in 6 of the 32 patients (18.7%) with positive C-reactive protein value ($p<.01$). Tocolysis was significantly more successful in the patients with negative C-reactive protein value.

In our study total maternal morbidity rate was 18.3%. The major cause of maternal morbidity was chorioamnionitis that later on developed into puerperal sepsis. Out of eleven patients of puerperal pyrexia, five were suffering from puerperal sepsis. All the five had positive cervical swab culture and elevated C-reactive protein level. Out of five, four had positive cervical swab culture for Esch. Coli and had sensitivity with antibiotics like Amicacin, Cefotaxim, Netlimycin and Gentamycin.

The remaining one had positive cervical swab culture for Klebsiella species and had sensitivity with antibiotics like Ceftriaxone, Cefotaxim, Cefoperazone, Neltimycin and Amikacin. All the five patients were treated with proper antibiotics and responded Well. Remaining six patients had normal C-reactive protein value. Out of six, three were suffering from Malaria (Plasmodium Vivax) and were treated with antimalarial drugs while

the other three patients with idiopathic fever became afebrile on second postnatal day without any treatment.

P. Hawry-Lyshyn, JE Milligen, S.Soldin, A Pollard and FR Papsin (1983) found maternal morbidity rate 16.7% while Anjana Devi and Reddi Rani found maternal morbidity 20.19% due to wound infection (7.69%) puerperal sepsis (8.65%) and urinary tract infection (3.85%). Our findings are also similar to Anu Mathur, S.S. Trivedi et al 1992). They found maternal morbidity 20% due to puerperal pyrexia.

Among 20%, 8.89% patients had positive cervical swab culture with elevated C-reactive protein level. Out of remaining 1.1% patients who had normal C-reactive protein value (but had puerperal pyrexia), 7.8% had malarial fever and responded to antimalarial treatment, 2.2% patients had mild hepatitis with low-grade fever and 2.2% patients became afebrile on 3rd postpartum day without any treatment.

Table-16 is showing co-relation of elevated C-reactive protein level in neonatal blood and neonatal morbidity in cases of premature rupture of membranes. C-reactive protein was raised more than 6 mg/litre in all the five septicaemic newborns (8.3%). Average value of C-reactive protein in septicemic newborns was 10.8 mg/litre. It was not raised (i.e. less than 6 mg/litre) in other causes of neonatal morbidity (like neonatal hyperbilirubinemia, neonatal respiratory distress syndrome and neonatal intraventricular haemorrhage). Our findings are in agreement with Peter J. Thompson, Anne Greenough. Edward Davis and Kypros

H.N. Nicolaides (1992). They suggested that elevation of C-reactive protein levels in newborns (i.e. $\geq$ 8 mg/litre) in the patients with preterm premature of membranes was associated with neonatal infections. Our findings are also similar to Anu Mathur. S.S Trivedi et al (1992). They found elevated C-reactive protein level in all the three (6.67%) septicaemic newborns but not in other causes of neonatal morbidity.

When we compared neonatal C-reactive protein levels with other tests as indicators of neonatal sepsis (e.g. total leucocute count, differential leucocyte count, and presence of polymorphs in gastric aspirate) C-reactive protein levels were raised in all the five septicemic babies and had accuracy of 100%. Leucopenia (less than 5,000 cells/cmm) was present in four out of five babies and had an accuracy of 80%. Presence of polymorphs more than 20/HPF in gastric aspirate of newborns was present in all the five septicemic babies and had accuracy had an accuracy of 100%.

On the other hand nonsepticemic newborns (n=53) did not have polymorphs in their gastric aspirates while only one non-septicemic newborn has leucopenia. Boyle et al (1978) have suggested that a count less than 10,000/cmm is associated with infection. Philip and Hewitt (1980) suggested the count less than 5000 cells/cmm to improve specificity of the test. Our findings are consistent with Singh et al (1987). They found that absolute leucocyte count less than 7,000 cells/cmm, 8 mg/litre and gastric aspirates having polymorphs more than 20/HPF had a sensitivity of 75% and specificity of 100% in diagnosing neonatal septicemia. According to Ajay Singh et al (1989) with total leucocyte count of

less than 5,000 cells/cmm had 62.96% sensitivity and 74.51% specificity in diagnosing neonatal septicemia.

The main causes of perinatal morbidity in study group are sepsis, neonatal hyperbilirubinaemia, neonatal respiratory distress syndrome and intraventricular haemorrhage.

Most common complications of premature rupture of membranes is neonatal hyperbilirubinaemia (15%) followed by neonatal sepsis (8.3%) and neonatal respiratory distress syndrome (5%) while the least common complication of premature rupture of membranes in perinatal morbidity is neonatal intraventricular haemorrhage (1.6%).

Total perinatal morbidity in our study group was 30%.

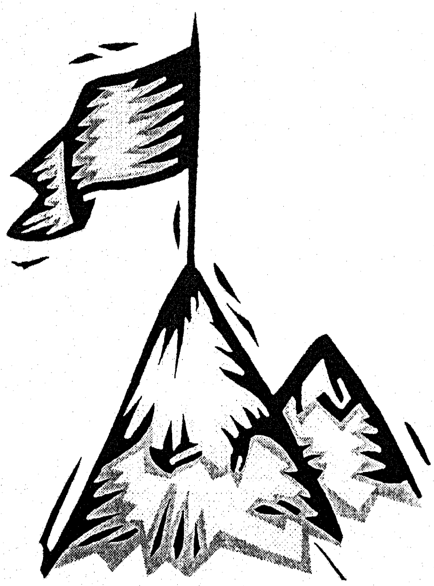
Our findings are in agreement with Anu Mathur, S.S. Trivedi et al (1992) who found total perinatal morbidity rate 22.1%, of which 6.6% was attributed to neonatal sepsis, 13.3% to hyperbilirubinaemia and 2.2 % to neonatal respiratory distress syndrome.

Total perinatal morbidity rate was 13.34% in our study, 50% of which was attribute to sepsis followed by 37.48% to neonatal respiratory distress syndrome and 12.5% to neonatal intraventricular haemorrhage.

Our findings are similar to Anu Mathur, S.S.Trivedi et al (1992). In this study total perinatal morbidity rate was 11.1%, 40% of its was attribute to sepsis.

Anjana Devi and Reddi Rani (1996) found perinatal morbidity rate of 4.8% mainly due to respiratory distress syndrome.

SUMMARY AND CONCLUSION



CONCLUSION AND SUMMARY

The study was conducted in the Department of Obstetrics and Gynaecology in collaboration with Department of Biochemistry in M.L.B. Hospital, M.L.B. Medical College, Jhansi since July 2002 to August 2003.

One hundred antenatal cases were selected from out patient department and in- patient department of Obstetrics and Gynaecology of M.L.B. Medical College Jhansi .The following samples were analysed.

1. Forty cases of normal pregnancy between 28-40 weeks of gestation taken as controls.
2. Sixty cases of confirmed premature rupture of membranes before 35 weeks of gestation taken as study group.

Any associated medical illness including infection, Rheumatoid arthritis or Systemic Lupus erythmatosus was excluded from the study.

Serial C-reactive protein determination was done in each patient. Blood sample was collected by venipunture.

C-reactive protein determination was done by using latex agglutination method with the help of C-reactive protein reagent kit.

The following conclusions were derived from present study: -

1. Highest incidence of premature rupture of membranes occurred in age group between 15-24 years (56.6%) and had mean age group of 25.15±5.1 years.

2. A high incidence of premature rupture of membranes was noted in low socio-economic group 60% as compared to only 6.6% in upper socioeconomic group.

3. Third gravida and forth gravida showed maximum incidence of premature rupture of membranes (40% and 20% respectively).

4. Most common gestational age for premature rupture of membranes in our study was 31-33 weeks (43.33%), followed by 34-36 weeks (36.67%) and the least incidence of premature rupture of membranes was in 28-30 weeks of gestational age group cases.

5. The highest number of patients (48.3%) had duration of premature rupture of membranes between 5-145 hours followed by145-290 hours (35.0%) and 290-435 hours (13.3%). Least number of patients (1.7%) had duration of premature rupture of membranes between 435-580 hours & 580-725 hours. Mean duration of premature rupture of membranes since delivery was 169.06 hours.

6. In study group total number of infants were 60 and percentage of live birth was 96.7% while only two babies were still born (3.3%).

7. Percentage of male newborns was 63.8% and percentage of female newborns was 36.67% in our study group. The percentage of male newborn was higher in study group as compared to control group.

8. C-reactive protein was elevated in all cases of clinical chorioamnionitis.

9. On comparing C-reactive protein levels with other laboratory tests as indicators of infection (e.g. WBC count, DLC count, ESR, maternal temperature, fetal heart rate) we found C-reactive protein level to be more sensitive (91.6%) but less specific (58.3%) in identifying clinical chorioamnionitis. C-reactive protein was elevated in all cases of clinical chorioamnionitis, suggesting a specific role in tissue regeneration and repair.

10. The correlation with hisotpahtological chorioamnionitis was much higher for C-reactive protein determination than for the other tests with respect to sensitivity (91.16%) and specificity (96.1%). It seems clear that histologic changes precedes the clinical manifestation of chorioamninitis, and in this case it is logical to assume that C-reactive protein is an early predictor of an inflammatory process. It was found to be raised at least

48 hours before the onset of signs and symptoms of clinical chorio-amnionitis.

11. The histologic presence of chorioamnionitis in 58.8% of the placentas and amniotic membranes in the present study further shed light on the incidence of subclinical infection in these patients, and it suggests that this may be a contributing factor in the etiology of premature rupture of membranes. This finding is consistent with other reports in the literature concerning this clinical condition. The high incidence of hisotpahtological chorioamnionitis which was found in our study group and the positive predictive value of elevated C-reactive protein for this condition make C-reactive protein determination a valuable tool for the conservative management of premature rupture of membranes.

12. The results of this study also show that many of the traditional laboratory tests relied upon to predict the development of chorioamnionitis are unreliable. Pregnancy affects the white blood cell count in a variable fashion. Physiologic stress and betamethasone administration significantly elevate white blood cell counts. Manual band neutrophil counts are highly subjective. The ESR is elevated in pregnancy because of its dependency on plasma protein characteristics and it also has a wide range of normal values. Only C-reactive protein determination accurately reflected chorioamnionitis with both high sensitivity and specificity. however, elevated C-reactive protein levels correlated better with histopatho-logical evidence of chorioamnionitis than

with clinical febrile morbidity. This reflects the physiologic role of C-reactive protein in the acute phase protein response to inflammation or tissue destruction, rather than the pathogenesis of febrile morbidity.

13. Positive CRP-values are associated with significantly lowered prolongation of pregnancy by tocolysis and subsequently lowered gestational age at birth. The mean hours to delivery for the patients with negative C-reactive protein value were 264.1 hours versus 85.87 hours for patients with positive C-reactive protein value ($p<.01$). Tocolysis was successful in prolonging gestation for 1week in 89.3% with a negative C-reactive protein value while tocolysis was successful only in 18.7% with positive C-reactive protein value ($p<.01$). Thus tocolysis was significantly more successful in the patients with negative C-reactive protein value.

14. Total maternal morbidity rate was 18.3 %. The major cause of maternal morbidity was chorioamnionitis, which later on developed into puerperal sepsis .In all the cases of chorioamnionitis cervical swab culture was positive and C-reactive protein levels were elevated.

15. Total perinatal morbidity rate in premature rupture of membranes was 30%. The main Causes of perinatal morbidity were neonatal sepsis, neonatal hyperbilirubinaemia, neonatal respiratory distress syndrome and intraventricular haemorrhage while most common complication of premature rupture of membranes in perinatal morbidity is neonatal

hyperbilirubinaemia (15.51%) followed by neonatal sepsis (8.6%) and neonatal respiratory distress syndrome (5.17%).

16. Total perinatal mortality rate was 13.34% of which attribute to sepsis followed by 37.48% to neonatal respiratory distress syndrome and 12.51% to neonatal intraventricular haemorrhage.

17. C-reactive protein was raised more than 6 mg/litre in all the 5 septicaemic babies (8.6%). It was not raised (less than 6 mg/litre) in other causes of neonatal morbidity (like neonate hyperbilirubin-aemia, neonatal respiratory distress syndrome and neonatal intraventricular haemorrhage). Average value of C-reactive protein in septicemic babies was 20.8 mg/litre.

18. When we compared neonatal C-reactive protein levels with other tests as indicator of neonatal sepsis (e.g. total leucocyte count, differential leucocyte count and presence of polymorphs in gastric aspirate of newborn). We found C-reactive protein level was raised (>6 mg/litre) in all the 5 septicemic babies & had an accuracy of 100%. While Leucopenia had an accuracy of 80% & Percentage of polymorphs less than 50% also had an accuracy of 80%. But presence of polymorphs more than 20/HPF in gastric aspirate of newborns also had an accuracy of 100%.

    Our results suggests that C-reactive protein may be reliable, early predictor of infectious morbidity and thus may be of benefit in the selective management of patients of premature rupture of membranes.

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